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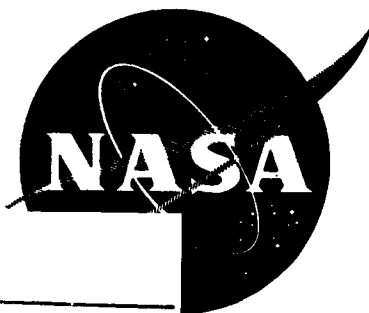
# MICROBIOLOGICAL FLORA OF HUMAN SUBJECTS UNDER SIMULATED SPACE ENVIRONMENTS

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SUBJECTS UNDER SIMULATED SPACE  
ENVIRONMENTS**

*PHYLLIS E. RIELY  
DIANE J. SHORENSTEIN*

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## FOREWORD

This is the final report of a study conducted at both the Aerospace Medical Research Laboratories, Wright-Patterson Air Force Base, and Republic Aviation Division of Fairchild Hiller Corporation, under contract AF33(615)-3255. This was initiated under Project 7164, "Biomedical Criteria for Aerospace Flight;" Task No. 716405, "Aerospace Nutrition," and completed under Project 6373, Aerospace Life Support; Task 637306, Aerospace Sanitation and Personal Hygiene." It was accomplished in conjunction with the National Aeronautical Space Administration (NASA), Manned Spacecraft Center, Houston, Texas under contract No. R-85, "The Protein, Water and Energy Requirements of Man Under Simulated Space Conditions." This contract, initiated by Dr. S. A. London, was completed under the direction of Dr. A. E. Prince, Biotechnology Branch, Life Support Division, Biomedical Laboratory, of the Aerospace Medical Research Laboratories. The research reported herein was started August 1965 and completed October 1966.

The authors wish to acknowledge the invaluable administrative assistance of Mr. Darrell Beard and the excellent technical contributions of Mrs. Fay Ames, Mrs. Charlotte Titus, and Miss Elisabeth Moss of the Republic Aviation Division of Fairchild Hiller.

The statistical section was prepared under the supervision of Mr. Herbert Jaffee and the authors wish to express their appreciation to him.

This report has been reviewed and is approved.

WAYNE H. McCANDLESS  
Technical Director  
Biomedical Laboratory  
Aerospace Medical Research Laboratories

## ABSTRACT

Aerobic and anaerobic microbiological studies were conducted on selected body areas of 11 human male subjects living under controlled conditions. Similar studies also were made on specific objects located in their environmental area. The data from these studies have provided information on microbial dynamics and bacterial levels, as influenced by various personal hygiene procedures and confinement. Microbial studies (both aerobic and anaerobic) of the fecal flora showed the influence of defined space-type diets. A statistical treatment of the data has helped to direct the formulation of personal hygiene procedures that should keep the bacterial populations within a numerically normal range for an individual. This analysis confirmed the importance of the groin and glans penis, as well as the axilla, as the most significant numerical indicator areas of microbial buildup. A detailed study of the predominating fecal anaerobes was conducted to classify these bacteria into recognized generic groups.

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## SECTION I

### INTRODUCTION

One of the most important conditions that must be investigated before extended space missions are undertaken is the effect such missions would have on the indigenous microflora of man. The degree of effect and how to use this information to establish realistic personal hygiene protocols for safeguarding the well-being of the astronaut must be determined also. For these reasons, study contracts were initiated by the Aerospace Medical Research Laboratory to determine the microbial flora of young, healthy, male adults. To eliminate extraneous influences (i.e., unrelated environmental factors), both the environment and the subjects were strictly controlled. This control allowed a valid evaluation of the complex microbial interactions among men, between men and their environment, and those within the man himself.

Many microbial forms contribute to the balanced indigenous microbial populations of any particular body locale. It is the maintenance of the balance of the resulting flora that may be a key to the health and well-being of the man. It is essential not to alter this flora by extreme changes in dietary regimen, or by the use of topical agents on the skin. If the intestinal microflora is altered by a dietary change<sup>(1)</sup>, the resulting flora may not be favorable to the health of the man. An additional factor to be considered is the difficulty of restoring the intestinal flora to its original stable balance. The same problems exist for the skin. The use of topical agents (or particular cleansing agents) often results in a selected microbial population<sup>(2)</sup>. This alters the immunological response of the body, since the protective mechanisms afforded by certain bacteria may be lost.

Microbial populations differ widely in different body areas. Bacterial forms which are "normal" to one area (for example, those of intestines) are not indigenous to another area (for example, the skin). One of the potential dangers in space travel is the transference of indigenous flora to another locale, where they could become pathogens.



On the skin of the host, the pathogen must compete with the resident flora for a specific habitat. This competition is influenced by many factors:<sup>(3)</sup>

(1) the bathing habits of the host, which include such specifics as the frequency of bathing, the kind and the method of applying soap, the temperature of the water, the length of time the soap is in contact with the skin before rinsing, the efficiency of the rinse, the actual pressure of toweling, to say nothing of the resident microbial population of either the wash cloth or the towel; (2) the variation in the perspiration levels, as well as the kind of perspiration (dependent upon the glandular source); (3) the pH of the skin; (4) the kinds and amount of clothing and their bacterial levels, materials (porous or nonporous), fiber content, as well as the degree of constriction of the garment; (5) the distribution of hair on the body, which is governed by sex, age, and racial differences; (6) the level of environmental contaminants; (7) the application and/or frequency of use of topical agents; (8) the variation in body temperature induced by the environment, clothing, or hygienic measures; and (9) the illusive factor of individual resistance or susceptibility, which may be the sum total of all these factors as well as the medical status of the individual.

The evaluation of the competition between bacterial forms in or on a host is complicated by transient shifts in microbial populations due to "tourist bacteria" and the effect of personal hygiene procedures.

To establish the significance of general trends and to deemphasize minute transient shifts or changes in microbial populations, the numerical data obtained during the study were treated statistically. By using this method, it was possible to define the time period when the bacterial levels became statistically significant. This basic information enabled the formulation of a realistic personal hygiene protocol for space missions.

## SECTION II

### MATERIALS AND METHODS

#### COLLECTION OF SAMPLES

The procedures for collecting samples from the body areas, feces, environmental, and miscellaneous areas are described for each class of samples.

##### Body Areas

Two swabs from each body area sampled were collected by subjects in either the Controlled activity Facility (CAF) or Life Support Systems Evaluator (LSSE) at 8:00-10:00 a.m. on specified days (Table 1). One swab was placed in 10 ml of Gall's broth plus cysteine for anaerobic culturing and one was placed in 10 ml of heart infusion broth for aerobic culturing. Collection was made by swabbing a specified area as follows:

Eye: Evert lower eyelid and swab conjunctiva gently, following contour of eyelid with swab.

Groin: Swab from front toward rear.

Axilla: Swab with care to get specimen from skin below hair area.

Throat: While depressing tongue, swab tonsillar area.

Mouth Area: Swab gingival margin adjacent to the last upper right molar.

Glans Penis: Swab specified area of skin of glans, or between glans and foreskin.

Ear: While pushing earlobe down and toward neck, gently swab external auditory canal with a circular motion.

Nose: While pushing the fleshy tip of the nose upwards, gently insert swab and rotate.

Umbilicus: Gently expose deeper folds of umbilicus by pulling upwards on surrounding abdominal tissue in order to swab all areas.

Anal Fold: Gently roll swab over area immediately adjacent to external anal sphincter.

Toes (Interdigital Spaces): Swab area between toes.

Scalp: Swab with a scraping motion within the area of hair growth.

Tongue: Roll swab from left to right on posterior portion of tongue.

Gingiva: Obtain samples from the appropriate areas with dental instruments.

For purposes of approximate quantitation each swab was considered to contain about 0.01 g of sample. This estimate was based upon intensive laboratory tests.

### Feces

Feces was excreted into plastic containers and samples were taken for culturing within 15 minutes after elimination.

### Environmental Areas

Aerobic cultures were made from several room areas, using two procedures:

Sedimentation plates of blood, MacConkey's, actinomyces agar, and phytone yeast were made from the following room areas by exposing the plates for 30 minutes.

- Table, fore (eating) and aft (games, etc.)\*
- Bed
- Floor, personal hygiene area

The following areas were swabbed. These swabs were placed in 10 ml broth and incubated aerobically.

- Communication equipment
- Personal hygiene seat

## PRIMARY CULTURING

### Primary Culturing of Microorganisms from Body Areas

#### Aerobic Series

The material on the swab collected by each subject from all designated body areas was emulsified in the 10 ml of broth into which it had been placed when collected.

---

\*One table only on Experiment XI

Tenfold serial dilutions in 4 to 6 tubes of trypticase soy broth were made depending upon the numbers of organisms expected to be present in the sample based on previous experience. The exact procedure for culturing is shown in Figure 1. The trypticase soy broth series was incubated aerobically and observed for growth at 24 and 48 hours. All cultures showing growth were examined microscopically. Aerobic plates were made on the media listed in Table 2 for each of the body areas by spreading 0.1 ml of broth from the most suitable dilution on the plate using a glass spreader. An additional blood agar plate was made in the same manner from the initial dilution. The aerobic count was obtained from a blood plate according to standard techniques.

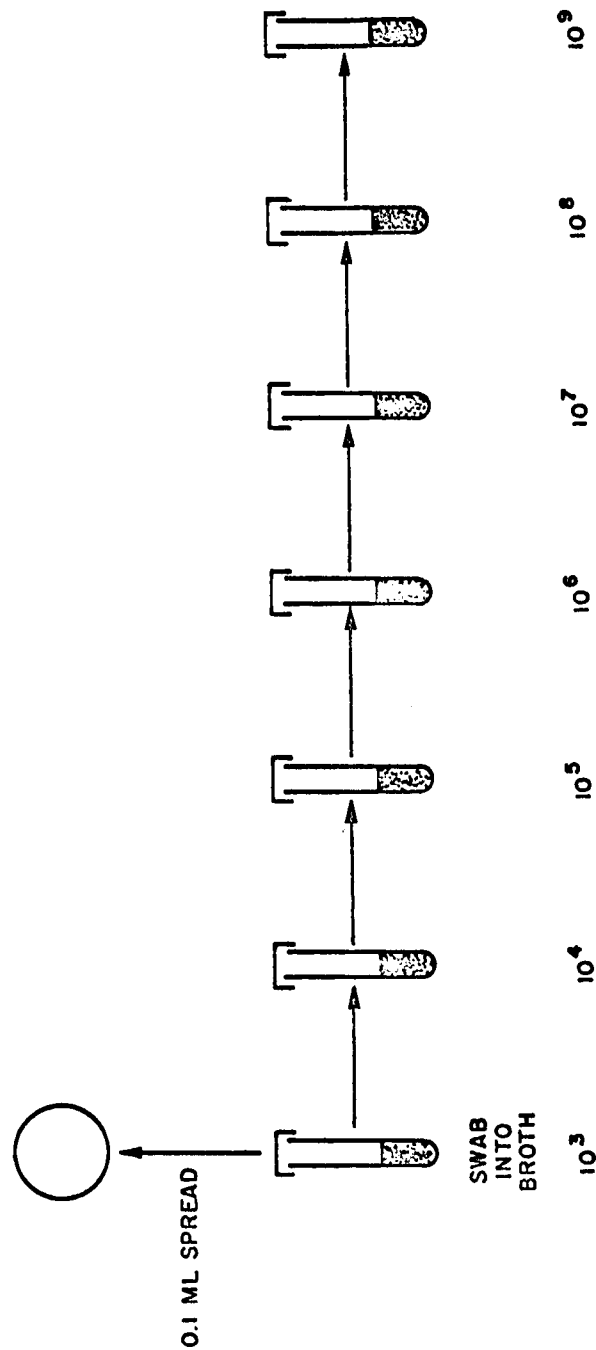
#### Anaerobic Series

The material on a swab from each body area (collected by each subject in the LSSE or CAF) was emulsified in 10 ml of broth. The sample was then serially diluted by tenfold dilutions depending upon the numbers of organisms expected to be found in that particular sample. The procedure, which is essentially the same as the aerobic method, is depicted in Figure 1. The cultures were then placed in an anaerobic jar, incubated at 37 C in an atmosphere of 10% CO<sub>2</sub>, and observed after 24 and 48 hours for growth. Agar shakes in Gall's agar, as well as slides, were made from the top dilutions showing growth. The agar shakes were then transported from the site of primary culturing to Republic Aviation Division's laboratories where the cultures were identified. In addition to the serial dilutions, anaerobic pour plates were made with 1.0 ml of the appropriate dilution from the throat, mouth, and glans penis samples using Gall's agar with cysteine. A blood agar plate and, where indicated, a chocolate agar plate were inoculated with 0.1 ml from the second dilution tube and spread over the surface of the plate with a sterile, bent glass rod. A pour plate of Rogosa's agar was inoculated with 1.0 ml from the appropriate dilution tube. These plates were incubated in the 10% CO<sub>2</sub> anaerobic jar.

#### Culturing of Fecal Microbes

##### Aerobic Series

The samples for the aerobic plates were taken from the anaerobic broth series. One-tenth ml from the third dilution tube was used as



Platings are dependent upon prior counts and change during the run. The counts resulting from these varied dilutions are changed and recorded as would appear on  $10^4$ .

Figure 1. Aerobic or Anaerobic Culture Series for All Body Areas

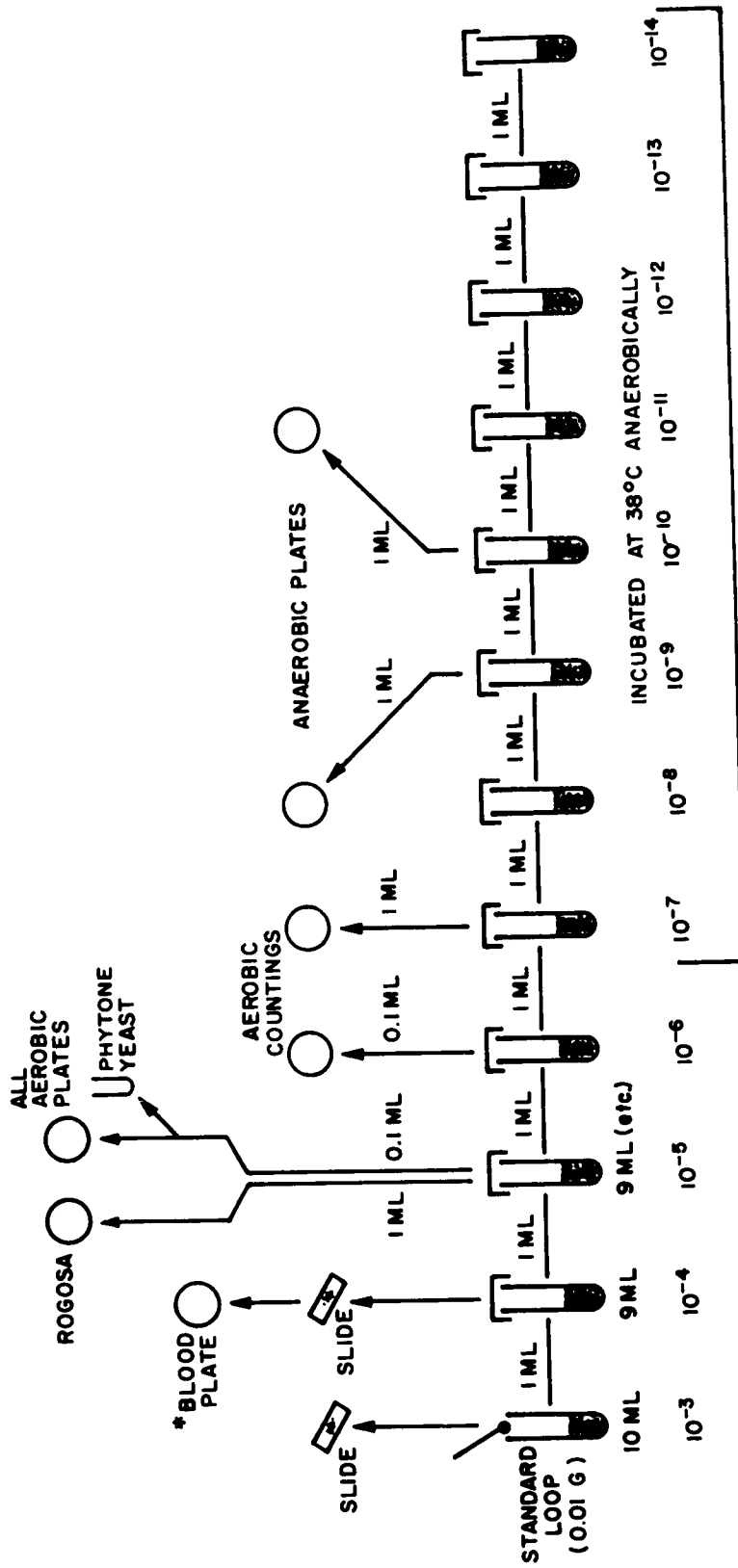
the inoculum for all aerobic plates, as well as the anaerobic blood plate. This was spread with a sterile bent glass rod upon the surface of the media. One-tenth ml from this dilution tube was also used as inoculum for a pour plate for the aerobic count. One ml from the third dilution tube was used as inoculum for Rogosa's pour plate.

### Anaerobic Series

The anaerobic broth series for the primary culture of the fecal sample was essentially the same as that used previously by Gall, et al. <sup>(4)</sup> for culturing rumen anaerobes, and which has been recently successfully adapted in the Republic laboratories to the culture of human feces <sup>(5)</sup>. This is a technique that can be adapted easily for work under field conditions. Figure 2 gives a schematic representation of the primary culturing technique, which is modified to culture from a standard loopful (0.01 gram) of freshly eliminated fecal material. Samples were cultured within 15 minutes of elimination.

The fecal material on the standard loop was placed directly into a tube containing 10 ml of Gall's broth prepared by adding 0.1 ml cysteine sodium bicarbonate solution. This tube was considered to represent roughly a  $10^{-3}$  dilution of the fecal contents. Serial dilutions were made into 11 additional tubes containing 9 ml of Gall's broth prepared as above by transferring 0.1 ml from the inoculated tube into the next tube, etc. The top 10 tubes were incubated in an anaerobic jar containing a 10%  $\text{CO}_2$  atmosphere until growth occurred. Observations for growth were made at 24 and 48 hours and at appropriate intervals thereafter. Growth usually appeared within 48 hours. These ten tubes were considered to approximate a dilution of the sample from  $10^{-5}$  to  $10^{-14}$ . No dilution blanks were used, as each tube containing broth acts as a dilution blank for the next tube in the series. One ml of broth from tubes 5 and 6 was used to make anaerobic pour plates by adding Gall's agar with cysteine bicarbonate solution.

The top three tubes showing growth were subcultured into agar shakes using Gall's medium to observe the anaerobic or aerobic character of the microorganisms and to preserve the cultures for transport, purification, and further study. Each culture was stained by Hucker's modification of the Gram stain and the slide was observed microscopically.



\* For additional identifications

Figure 2. Anaerobic Dilution Series (Feces)

Blood plates were made from the  $10^{-3}$  and  $10^{-4}$  dilution of the fecal sample by the same technique as the aerobic plates from the other body areas and were incubated at 37°C in the same manner as the anaerobic broth series; i.e., in 10% CO<sub>2</sub> atmosphere in an anaerobic jar. Growth was recorded after 24 hours and the plates were treated in the same manner as the anaerobic blood plates described below.

#### Environmental Areas

The sedimentation plates made from the several room areas indicated previously were exposed for 30 minutes, incubated at 37 C, and observed for growth at the end of 24 hours. The swab cultures taken from the environmental areas were placed in broth and incubated aerobically at 37 C. Smears were made of all broths that grew.

### SECONDARY CULTURING

#### Aerobic Series

All the cultures from the petri dishes incubated aerobically and anaerobically from all body areas, feces, environmental areas, and miscellaneous items were returned to the Republic Aviation Division's laboratories where selected colonies were picked into broth. Cultures picked from the anaerobically incubated plates were incubated in the CO<sub>2</sub> incubator while all other colonies from the anaerobic plates were processed by the usual aerobic methods. The cultures were smeared, stained, observed microscopically, separated according to morphological types, and processed according to the schema if applicable.

#### Staphylococci\* and Micrococci

- Mannitol salt agar
- All positives confirmed with coagulase test
- Phage typing on selected cultures

---

\* The identification of the staphylococci on Experiments IX, X and Xa are being carried out under separate contract by personnel from the Miami Valley Hospital Research Department, Dayton, Ohio. The results of this work are not included in the overall summary and tables.

Identification of staphylococci on Experiment XI was done by Republic Aviation Division of Fairchild Hiller.



### Streptococci<sup>\*</sup>

- Alpha hemolysis
- Beta hemolysis
- Gamma hemolysis
- Differential sugars
- Typing
- Temperature
- Salt tolerance

### Pneumococci

- Pneumococcus broth - bile solubility

### Haemophilus

- Isolated strains identified with typing antisera

### Neisseria

- Sugar screen test
- Oxidase test

### Lactobacillus

- Culture and morphology in Rogosa's medium
- pH in glucose broth
- Ecology

### Gram-positive Rods

- Loeffler's
- Morphology
- Gelatin
- Sugar screen
- Hydrolysis of starch
- Detection of hyphae (Actinomycetales)
- Tellurite
- Catalase
- Hemolysis on sheep blood
- CO<sub>2</sub> requirement
- Litmus milk

---

\* Experiments IX, X and Xa - Work performed by A. West, Research Microbiologist, Aerospace Medical Research Laboratories, Wright-Patterson Air Force Base, Ohio.

Experiment XI - Identification done by the Republic Aviation Division of Fairchild Hiller.

#### Gram-negative Rods

- TSI
- Indol
- Methyl red
- Voges-Proskauer
- Simmon's citrate
- Urease
- Nitrate
- Motility
- Gelatin
- KCN
- Phenylalanine
- Cytochrome oxidase (on all alkaline over alkaline TSI's)
- Typing antisera (shigella, salmonella, E. coli, klebsiella)

#### PPLO

- Dienes' stained agar technique

#### Fungi

- Phytone yeast media
- Wet mount
- Lactophenol cotton blue
- Corn meal agar
- Fermentation series when indicated

#### Actinomycetales

- Actinomyces media
- Morphology in culture, smears and wet mounts
- Biochemical series

#### Spirochaetes

- Darkfield when indicated
- Vincent's stain

#### Protozoa

- Identification by selective stains

#### Anaerobic Series

##### Body Areas

The agar shakes made from the dilution series and the colonies picked from the Brewer plate (when made) were separated into two groups depending upon the degree of anaerobiosis. The obligate anaerobes were processed in the same way as the fecal anaerobes described below with the exception that many of

the cultures, particularly from the mouth, gingiva, throat, and glans penis were identified from Bergey's manual<sup>(6)</sup>. The facultative anaerobes were grouped according to morphology and were processed as described in this section under "Secondary Culturing - Aerobic." A morphological and biochemical key was established consisting of the results of the screen tests from the most frequently occurring fecal anaerobic cultures and was designed to group similar bacteria. Each different screen test pattern was assigned an FA, FN, or GD number. The FA and GD types were used to designate obligate anaerobes and the FN types to designate facultative anaerobes (see Table 3).

### Feces

The agar shakes from the top three tubes of the cultural series were processed in the following manner. The agar shake cultures were transferred to Gall's broth plus cysteine and incubated anaerobically until growth occurred. Gram stains were made and, if the cultures were pure, they were immediately screen tested as described below. Cultures showing two or more distinct morphological types of bacteria were purified by plating using the following anaerobic technique. A diluted drop of the impure broth culture was spread on a bed of Gall's agar which was then covered with a layer of Gall's agar with added cysteine. The plates were incubated anaerobically in a Torbal jar with hydrogen and 10% CO<sub>2</sub>, and discrete colonies were picked. Selected colonies on the anaerobic Brewer dishes originating from tubes 5 and 6 were picked and treated like the subcultures from the agar shakes as described above. The physiological studies of the pure cultures isolated from the feces included the following screen tests:

- Gram stain to observe morphology
- Final pH in 0.1% glucose broth
- Fermentation of the following sugars in Gall's media with glucose omitted (glucose, sucrose, lactose, dextrin - sugars added at 0.1% level aseptically after autoclaving)
- Growth in Gall's broth with no carbohydrate added
- Liquefaction of 12% gelatin in Gall's medium minus carbohydrate
- Growth and reaction in litmus milk (to which 0.05% bovine albumin and 0.1% of peptone have been added)
- Growth in agar shake containing Gall's medium

All media contained bicarbonate and all media except the agar shake contained cysteine to produce an Eh of about -200 mv. The results of the screen tests on each anaerobic culture were compared with a "key."

## GALL'S MEDIUM

Purpose: Anaerobic culturing

<u>Formula:</u>	Peptone C (Albimi)	1.0%
	Peptone S (Albimi)	1.0%
	Beef Extract (Difco)	1.0%
	Yeast Extract (Difco)	1.0%
	$K_2HPO_4$	0.1%
	$KH_2PO_4$	0.1%
	Glucose	0.1%

Technique: Make up to 100 ml with distilled water and tube in 9 ml amounts (pipetted for exactness of dilution) and sterilize exactly 10 minutes by autoclaving. Immediately before use, add aseptically 1 drop of sterile 10%  $NaHCO_3$  and two drops of 10% cysteine-bicarbonate solution. \*<sup>3</sup> This gives a pH of approximately 6.8 and an Eh of approximately - 200 mv. Add 1.5% agar to the above when agar is needed for shakes and plates. This is done when originally making the media. In agar omit cysteine except where noted otherwise. To all broth and agar media add 0.05% of bovine serum.

### \*10% Cysteine-Bicarbonate Solution

20 g Cysteine Hydrochloride  
100 ml 1N NaOH  
7%  $NaHCO_3$

Add the cysteine hydrochloride to the NaOH, giving an approximate pH of 7.0.

More or less NaOH will be needed depending on the particular batch of cysteine hydrochloride.

To 4 ml of this solution (15% cysteine) in a test tube, add 2 ml of 7%  $NaHCO_3$ . Seal with melted vaspar. Autoclave at 15 lb for <sup>3</sup>10 minutes.

## GALL'S GELATIN (i. e. 12%)

Purpose: The use of gelatin in culture media for studies of gelatinolysis (elaboration of gelatinolytic enzymes) by bacteria.

<u>Formula:</u>	Bacto tryptone	10 g
	Bacto peptone	10 g
	Bacto yeast extract	10 g
	Bacto beef extract	10 g
	Monobasic potassium phosphate	1 g
	Dibasic potassium phosphate	1 g
	Serum	1 cc
	Gelatin	120 g

### SECTION III

#### EXPERIMENTAL RESULTS

##### OBJECTIVE

The purpose of this study was to investigate the composition of the indigenous biological flora of 11 human male subjects under controlled experimental conditions; and to study the effects of diet upon the fecal flora. In addition, the bilateral microbial character of the groin area was studied both qualitatively and quantitatively. The endemic situation in both the Controlled Activity Facility (CAF) and the Life Support Systems Evaluator (LSSE) was evaluated.

In addition, the number of typable strains of E. coli present in eight fecal specimens (standard methods) was determined. This was done to substantiate results obtained in previous studies where a large number of the E. coli gave specific serum types. Gingival samples were obtained from 10 subjects to determine if this was a significant area microbiologically, either quantitatively or qualitatively.

##### DESCRIPTION

During three different experimental periods, 11 subjects of normal health were confined in the CAF and the LSSE. In the first experimental period, four male subjects of normal health were confined in the CAF for two weeks, transferred to the LSSE for 15 days, during which time two of the four subjects were suited in the MA-10 space suit, and then returned to the CAF for the final 14 days of the experiment. The subjects were sweat tested 10 times during the experiment. Each sweat test required that the subject be scrubbed by the monitor two times with soap, rinsed three times, then rinsed two times the following day.

In the second experimental period, three subjects (all Air Force personnel) were confined in the CAF for 3 days, in the LSSE for 15 days with the door sealed, and in the LSSE for 3 days with the door unsealed. Three men wore the suits for the first 7 days in the LSSE. The use of suits was discontinued for the remainder of the experiment because of difficulty with the blower mechanism of the suits.

In the third experimental period, four subjects were confined in the CAF for 45 days followed by 10 days in the LSSE and then 5 days in the CAF. One subject wore an Apollo suit and one a Gemini suit while in the LSSE.

#### EXPERIMENTAL DESIGN

The design of the various experimental periods is shown in Table 1. During the first period the A Areas\* were sampled 11 times and the B Areas\* 3 times. The feces were sampled 11 times. During the second period, the A Areas were sampled 9 times and the B Areas 3 times. The feces were sampled 6 times from Subjects A and B and 3 times from Subject C.\*\* In the third experimental period, the design of the experiment was radically altered. The body areas were sampled 26 times. The areas sampled were the left and right groin and the gingival area. The feces were sampled 15 times from Subjects 41 and 44, 13 times from Subject 42, and 14 times from Subject 43.

In evaluating the results, it was necessary to consider the variations present during the different periods. In Period 1, the sweat tests may well have influenced the total bacterial levels, while in the third experimental period the variation in dental hygiene must be considered in evaluating the gingival results. While in the CAF, the dental hygiene consisted of brushing after every meal on days 1 through 15, no brushing on days 16 through 20, and while in the LSSE, brushing once a day. One subject did not brush his teeth until day 43 and subsequently brushed more frequently than the experimental design indicated. In addition, during the third experimental period the subjects were not strictly confined in the CAF during days 1 through 5. There was no screen on the filtering system during days 26 through 29. On day 35 the subjects left the CAF for altitude indoctrination.

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\*A and B Areas are described in Section II.

\*\*Subjects A, B, and C were Air Force personnel as opposed to subjects 1 through 44, who were civilian employees.



## RESULTS

The quantitative results of the environmental sampling are shown in Table 4. While minor fluctuations are present, there appears to be general rise in the level of bacteria proportional to the time of occupancy. The types of organisms isolated from the varied plates used in the sampling procedure (Section II) are shown in Table 5. In addition, swabs taken from selected room areas were cultured to indicate any possible interchange between man and the environment.

In the first experimental period, the isolation of Enterobacteriaceae on the bed, aft table, and the floor of the personal hygiene area indicates the necessity for more strict personal hygiene procedures to maintain a satisfactory level of sanitation. Results from the data of the second experimental period were the same and, in addition, enteropathogenic E. coli were isolated from the bed. In the third experimental period, Staphylococcus aureus was recovered from the environmental areas as well as from the subjects and the importance of transference of this organism will be presented in Section VI.

To determine the possible effect of simulated space conditions on the number of bacteria present on body areas, quantitative data were obtained from the bacterial samples. These total bacterial counts by body areas are shown in Table 6. During the first experimental period, the rather wide cycling in counts may be in part attributed to the frequent sweat testing. However, the counts on Subject 40 were generally lower than the other subjects, probably illustrating individual variation. The variations in anal counts are merely a reflection of personal hygiene procedures and individual performance. During the second experimental period, the same cycling appeared. The appearance of Enterobacteria on the axilla seemed to coincide with wearing the space suit and was followed by a gradual decline of these organisms and eventual disappearance of these bacteria after the suit was removed. During the third experimental period the wide cycling in gingival counts may reflect variations in the vigor with which the dental hygiene procedures were practiced or in the effectiveness of the sampling.

During the third experimental period, both the left and right groin areas were sampled 26 times (Table 7). E. coli was isolated only from the right groin

on Subject 41. Subject 42 carried E. coli with somewhat greater frequency than Subject 41, but only on the left groin, and Subject 44 frequently carried gram-negative rods (mostly Aerobacter species) on either the left or right groin. The bilateral recovery of fungi is more consistent since Subject 41 carried Trichosporon on both the left and right groin in the majority of the sampling periods. The qualitative differences between the right and left groin are both apparent and surprising since, if the counts are averaged for each subject, the quantitative results are very similar.

One of the most interesting studies was in the relationship of corynebacteria to staphylococci in the various body areas (as shown in Table 8 ). During the first period, corynebacteria predominated or were of the same order of magnitude as the staphylococci on the groin of all subjects. The only exception occurred in Sampling Period 4 where there was a dramatic drop in the count. Subject 40 illustrates individual variation, since his incidence of corynebacteria was low in relationship to staphylococci. During the second experimental period, Subjects B and C displayed the same relationships. The distribution of the varying strains of corynebacteria is shown in Table 9 . C. pseudodiphtheriticum appeared to predominate on the nose of all subjects while the other body areas showed one major strain and other strains sporadically isolated. Very often the groin and glans penis carried the same strain at a given sampling period. Table 10 shows the biochemical reactions upon which the patterns for the differentiation of the corynebacteria are based.

During the entire study, PPLO\* were recovered only from Subject 37. They were recovered from the tongue at the first sampling period and from the gingival area on the ninth sampling period.

Special actinomyces media were used in the sampling procedure. Table 11 shows the recovery of actinomyces and nocardia during these experimental periods. The appearance of these microorganisms seemed to be sporadic and the indigenous stature is questionable. Various members of the family Bacilliaceae were recovered throughout the experimental periods, and while charted, are felt to be air contaminants rather than members of the indigenous microbial population of the men. Lactobacilli are shown on Table 12. The low frequency of isolations in certain subjects was surprising. In particular, the lack of correlation of isolations from

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\*Mycoplasmataceae

the throat and feces of Subject 37 was surprising, since the literature indicates that lactobacilli are rarely found in the feces without concomitant presence in the throat.

During the first experimental period, neisseria was prevalent, with isolations from the throat and tongue of Subject 37 at all sampling periods and from Subjects 38, 39, and 40 at a majority of the sampling periods. Gaffkya was isolated from the throats of all subjects as well as from the tongues of Subjects 38, 39, and 40. During the second experimental period, a gaffkya-like organism occurred on Subjects A and B and, in addition, neisseria was prevalent on the tongue, throat, and gingival areas of these men (Table 13).

Table 7 shows the occurrence of enterobacteriaceae and related organisms. During the first experimental period, the spread of these organisms to the groin and glans penis is apparent. Noteworthy is the presence of proteus which was routinely isolated on the glans penis of Subject A and once on Subject B. Since this organism is known to be capable of causing serious urinary tract infection, this finding should be carefully considered in relation to use of any commonly shared urine transport device. The bacterial recoveries on Subjects 41, 42, and 43 were generally unremarkable with the exception of finding an occasional enteropathogenic type of E. coli on Subject 41. Subject 44, however, showed an interesting pattern of carrying proteus until the seventh fecal sample at which time the enteropathogenic coli type 026:B6 was isolated and subsequently recovered with great frequency throughout the rest of the experiment. The proteus did not reappear. (The seventh fecal sample was obtained at the end of the time on the contingency diet.)

The identification of staphylococci during the first two experimental periods was the responsibility of another agency. Since no data has been received from this group, only the data from the last experimental period during which the staphylococci were identified by this Microbiology Department will be discussed. The potential pathogenicity as indicated by phage typing was performed by Dr. John Blair, Head of the International Committee on Phage Typing at Roosevelt Hospital in New York City.

The reported isolation of phage typable strains of Staphylococcus aureus from the environment of the CAF is interesting from several viewpoints. Prior to this experimental period, efforts were made to clean (from the microbial viewpoint) the CAF using a bactericidal solution to scrub down all exposed areas and a spray for hard-to-reach areas. At the beginning of this particular experimental period (third), the CAF was not cleaned. After 6 days the atmosphere was sprayed with water to sediment particulate matter and the floor was washed with the same bactericidal solution. Transmission through air is a common fact and since surface contamination may be rendered airborne by numerous physical activities, the presence of these strains of Staphylococci aureus is still unusual since recovery of viable staphylococci is usually limited to 12 hours exposure at 50 C and 86% relative humidity<sup>(7)</sup>. The ability of Staphylococcus aureus to spread in a community is a reflection of its temporary ability to withstand drying.

The original infective source is not obvious, but it is assumed the source was human and may have been one of the monitoring personnel. Although phage patterns were isolated repeatedly as shown in Tables 14 and 15, from both the room areas and subjects in particular, the 52/52A/80/81 complex was isolated at 19 of the 26 sampling periods. It was first isolated from the floor in the personal hygiene area at the second sampling period, on the fifth sampling period from the table, and by the seventh sampling period was isolated from the gingiva of Subject 43. It then appeared in the feces and gingiva of Subject 44, and on the bed of Subject 43. Type 80/81 and its closely related types are responsible for many outbreaks of infection and have a great tendency to become resistant to penicillin and other antibiotics. The co-actions occurring when attempting to implant Staphylococcus aureus are poorly defined, and to at least some extent are dependent upon resident strains of other microorganisms - in particular Staphylococcus epidermis. Phage type 3B/3C was isolated about the midpoint of the experiment and was recovered first from the gingiva of Subject 42 and subsequently from his nose and many environmental areas. It was never isolated from the other subjects. Phage type 47/53/54/75 was isolated only from the environmental areas and was present throughout the experiment.

The occurrence of fungi on body areas is shown in Table 16. During the first experimental period, members of the Candida species were isolated from all subjects. Candida albicans was isolated only on Subject 40. Trichosporon was isolated on the ear and groin of Subject 38, on the toes of Subject 39, and glans penis of Subject 40. With the exception of T. rubrum on the toe of Subject 37 and T. tonsurans on the scalp of Subjects 37 and 40, the molds isolated were considered to be saprophytes. During the second experimental period, various Candida species were recovered from all subjects. Subject B carried C. guilliermondii exclusively, while C. albicans was found on the other subjects. The molds isolated were considered to be normally occurring saprophytes. During the third and longest experimental period, candida was recovered only four times. C. albicans was recovered from the feces at three different sampling periods and candida from the gingiva at one sampling period. Trichosporon was prevalent during the entire experimental period on Subject 41, but no permanent transfer occurred since it was isolated only from the groin of Subject 43 at one sampling period. Rhodotorula occurred sporadically in the feces of Subjects 41, 43, and 44. Note Subject 42 had only one isolation; cladosporium being found on the right groin at one sampling period. The environmental areas supported the usual common saprophytic inhabitants.

During a prior study<sup>(8)</sup> typable strains of E. coli were recovered from over 50% of the samples. Since this greatly exceeds the 2-5% occurrence of typable strains in the normal population<sup>(9)</sup>, greater emphasis was placed upon this identification during the present study. All coli occurring on MacConkey's plates in the range acceptable to standard methods<sup>(10)</sup> were identified at eight sampling periods. These results are shown in Table 17. Each colony was tested and those not conforming with standard identification were grouped and identified as patterns (Table 18). Those E. coli designated NT (no type) were tested with E. coli polyvalent A and polyvalent B serum and were found not to type with either.

The various patterns found may well be intermediates in the coli-aerobacter groups. According to Edwards and Ewing<sup>(11)</sup>, "Although there is always the tendency to think of the established groups as distinct entities, it should be kept in mind that not only do many intermediate strains exist, but there are many intergroup

relationships among typical strains of the various groups." Only on Subject 41 was there a definite shift in the type of coli present as the experiment progressed. He entered with a coli flora consisting exclusively of nontypable organisms, but by the sixteenth sampling period, greater than 50% of the coli isolated were of the enteropathogenic type 0125:B15. It may have been the changing of diets and ensuing unstabilized condition in the intestinal tract which allowed a minor organism in the flora to become predominant. On Subject 44, only one plate was analyzed at Sampling Period 16. Of the 75 colonies studied, 59 were typable E. coli Poly A 026:B6.

The dynamics of microbial growth, particularly in mixed cultures, are often surprising. The broth dilution series from which platings (at the appropriate dilution) to differential media were made, were incubated aerobically. On differential media and on blood plates, the corynebacteria usually predominated over the staphylococci; however, when these organisms grew together in broth cultures (Table 19), the staphylococci often outgrew the corynebacteria. This growth pattern may account for reports by some investigators on the predominance of staphylococci on the skin of the subjects they tested. However, the predominance of these organisms in a broth culture may be due either to their numerical superiority or to their production of an inhibitory substance which limited the growth and reproduction of corynebacteria.

Identification of the aerobic microorganisms recovered from fecal cultures is presented in Table 20. The specific identification of the gram negative rods is reported in Table 7, and the corynebacteria recovered during experiments X and Xa are charted separately, by strains, in Table 9. The occurrence of staphylococci was consistent in certain subjects, as was the appearance of Streptococcus viridans, and probably represents individual variation.

The estimated aerobic bacteria per gram of feces is shown in Table 21. While there is a wide fluctuation depending on the man and the sampling period, it is all within the range of 1 million to 100 million bacteria per gram. This contrasts with the anaerobic growth (Table 22) which indicates a minimum count in the billions and frequently a count two logs greater.

During the second experimental period 58 representative samples of all available diets were analyzed microbially. The results of this study are recorded in Table 23. Further identification of the organisms found on the primary plating was accomplished using the appropriate special media and biochemical tests. All mannitol positive staphylococci were tested for coagulase activity. There were no GD type anaerobes recovered. This eliminates these foods as a source of those anaerobes in the digestive system of the subjects. Two anaerobes which resembled FA-8 were recovered. The aerobes recovered, while not being of the "food poisoning" type of organism, should be considered as to their effects on food deterioration and their contribution to bad taste, odors, and changes in texture.

Obligate anaerobes recovered from body areas are shown in Table 24. The sporadic recovery of obligate anaerobes from certain body areas emphasizes the transient nature of the particular strains isolated. The gingiva and throat of the subjects support a true obligate flora with veillonella being predominant. Peptococci were found recurrently on both the groin and gingiva. In addition, they have been recovered from the glans penis and the anus, areas contiguous to the groin. Table 25 shows the repeated isolations of peptococci during the third experimental period.

Obligate anaerobes recovered from the feces are shown by subjects in Table 26 and by sampling period in Table 27. The results of these experimental periods are summarized in Table 28 and are compared with the data obtained from a study for NASA<sup>(12)</sup> in which the subjects were not on a defined diet or confined, and with the data obtained from another study<sup>(13)</sup> in which the subjects were on a defined diet and were confined (Table 29).

## SECTION IV

### STATISTICAL TREATMENT OF EXPERIMENTAL DATA

#### STATISTICAL APPROACH

This section describes the statistical evaluation and comparison of the data obtained from 1000 samples taken from specific preselected body areas of 20 human male subjects. In addition, the areas inhabited by the subjects were sampled 95 times.

For the numerical counts of the bacterial samples taken from the body areas, the mean, median, mode, and the standard deviation of the mean were calculated (Table 30). This basic information was used in the further analysis of these data.

The arithmetic mean was used because it was desired to obtain the measure of central tendency having the greatest efficiency and because it was required to compute the standard deviation and Students t ratio. The median was used to determine the midpoints of the numerical distributions. The magnitude of the extreme values, therefore, was of no significance to this median, since it only divided a number of items into two equal groups.

The standard deviation was used to compute the critical ratio (Students t ratio) and other statistics. The standard deviation is computed by taking the quadratic mean of the deviations from the arithmetic mean of the values. It is thus the root-mean-square of the deviations from the arithmetic mean.

#### T TEST

The testing of statistical hypotheses generally involves comparisons of numbers or statistics to determine the degree of difference between them and to ascertain whether a difference of this magnitude could be due to chance.



In testing the significance of differences, the null hypothesis is a useful tool. It assumes that the true difference between two values is zero -- or that the differences observed are normally distributed around zero. One can then compare the actual difference with the hypothesized zero difference to determine if the difference is significant statistically. In so doing, the null hypothesis can be rejected and it can be said that the differences observed are not due to chance.

Whether or not a difference is statistically significant depends on the probability of a certain value occurring due to chance. For biological data, significance at the .05 level is considered valid. That is to say not more than five times in one hundred could a difference as large as the size measured be expected due to chance.

After the mean and standard deviation of the mean were computed for each group of subjects at each point in time selected, it was then possible to test the significance of the difference between any two groups by employing the critical ratio (t test). For this test, the number of samples used from each body area was as follows:

• Anal area	112
• Axilla	193
• Groin	180
• Glans penis	125
• Gingiva	32
• Interdigital spaces (foot)	76

The  $t$  value is equivalent to the difference of the means divided by the standard error of the difference or:

$$t = \frac{\bar{X}_1 - \bar{X}_2}{\sqrt{\frac{\sigma_1^2}{N_1} + \frac{\sigma_2^2}{N_2}}}$$

$\bar{X}$  = Mean of X  
 $\sigma$  = Standard deviation  
 $N$  = Number of samples

The  $t$  value thus obtained can then be compared with a table indicating significance at various levels.

The null hypothesis was first tested at the baseline versus postevaluator period, and baseline versus 25 days into the experiment for each body area. If the difference was statistically significant at either of these points, intermediate intervals from the baseline data were tested, to indicate, as closely as possible, the period of time required for the bacterial count to build up to significantly higher levels.

From the table below it can be seen that on the axilla, an increase considered to be significant occurred by the 14th to 15th day of the experiment as well as between the 15th and 25th days. The difference (increase) between baseline and postevaluator period is significant. Between day 25 and postevaluator period the difference was not significant, indicating that the buildup in numbers of micro-organisms was maintained.

On the groin, the increase in numbers did not become significant until the 22nd to the 23rd day. The baseline numerical values versus those of the post-evaluator period were significant. Again, there was no significant difference in count between the 25th day and the postevaluator period, indicating that the buildup was maintained.

SIGNIFICANCE OF CHANGES IN BACTERIAL COUNTS (T-TEST)

Day	Axilla	Groin	Anal	Toe	Glans Penis	Gingiva
11-12	-	-	*	*	*	*
14-15	+ (.02)	*	-	*	*	*
17-18	+ (.01)	-	-	*	-	*
22-23	+ (.01)	+ (.05)	*	*	-	*
25	+ (.01)	+ (.01)	-	-	+ (.05)	-
Base vs Post	+ (.05)	+ (.01)	-	+ (.01)	*	-
25 vs Post	-	-	-	-	-	-

- Not significant  
\* No data

On the glans penis, significant buildup occurred at the 25th day, and was maintained, following a pattern similar to that of the bacterial buildup on the groin.

The increase of bacteria between and under the toes was significantly higher at the end of the experiment than at the beginning, but was not significant by the 25th day. Because the interdigital spaces were sampled at widely varying times in different experiments, it was not possible to pinpoint the precise time when the bacterial counts became significantly higher.

On the gingiva, although buildup occurred, there is no time at which the number of organisms increased to a level statistically significant above that of the baseline, indicating the presence of a homeostatic, or self-limiting factor or factors, in the mouth.

On the anal area, there was no sustained increase, as evidenced by the lack of significant difference between baseline and postevaluator counts. This was to be expected, since the anal area was subject to periodic wiping.

Applying the  $t$  test to the environmental area results did not indicate that the fluctuation reached statistically significant levels, although obvious increases occurred. These increases can be more clearly evaluated from the graphic presentation (Figures 3 through 13).

This study indicated that the groin, axilla, and possibly the glans penis, are the most significant indicator areas of bacterial buildup and should be selected for microbial monitoring. In addition, the study determined the length of time required for increases in the bacterial levels on these areas to become significantly higher than those of the baseline counts. Using this information, it should be possible to keep these counts within normal variation by selective washing at predetermined intervals. For example, the study results show that if the axillae were cleansed at least every 15 days, and the groin every 22 days, acceptable limits should be maintained.

## GRAPHIC PRESENTATION

After collating the data and performing the t test, it was possible to present the data thus obtained in graphic form. Each area was graphed showing the number of bacterial colonies recovered versus the number of days into the experiment. This shows the dynamics of the changes in bacterial populations as opposed to the statistical presentation of the t test, and indicates smaller variations that occur within the significant range. It also makes it possible to compare the data curves for the various body areas and to superimpose these on the environmental area curves.

The significant points of interest from each curve include the following:

Graphing of the data from the axilla (Figure 3 ) indicates a sharp rise in the numbers of bacteria between the 12th and 15th day and an even more marked increase to a peak between the 25th and 38th day, although the rise in bacteria, as shown by the t test, was significant by the 14th and 15th days.

The groin composite (Figure 4) indicates a somewhat greater fluctuation with a cycling effect apparent in the rise to day 14, a drop at day 23, a sharp rise by day 25, a slight plateau and then a rise to a sharp peak by day 38.

For the glans penis (Figure 5 ), graphing indicated a much lower overall count and the absence of cycling, although there was a plateau between days 12 and 20 and then a single sharp rise to a peak by day 28.

In the anal area continuous fluctuation is apparent (Figure 6 ), which further supports the evidence that there was not a sustained significant increase in this area. However, the overall counts, even when in regression, never fell to the original baseline level.

On the gingiva (Figure 7 ), as with the anal area, there was a continuous fluctuation but in this area the counts returned to their original levels. Since a measure of oral hygiene was employed, this was an expected result.

A comparison of the graphs on the axilla, groin, and glans penis indicates a general overall similarity, with the sharpest rise in bacterial counts occurring on the axilla and groin between days 25 and 35 and on the glans penis between days 22 and 30. This lends further evidence to the premise that these are buildup and key areas and should be monitored.

Both the anal and the gingival areas, which are similar by their continuous fluctuating levels of bacteria, indicate a numerical peak at day 15. However, as indicated by the *t* test, in neither case was the numerical difference significant.

Data from the environmental areas were first graphed by each experiment and then a composite, averaged graph of these data was constructed.

The individual graphs show widely varying values. In experiment V (Figure 8), the highest counts were obtained from the bed, reaching a maximum at day 22. In experiment VI (Figure 9), the highest counts occurred on the aft table (with the exception of the floor personal hygiene area at the beginning of the stay in the LSSE), reaching a maximum at day 28. In experiment VII (Figure 10), the overall counts were lower with all areas reaching a maximum contamination at day 21 while the subjects were in the LSSE.

In experiment VIII (Figure 11), the pattern of contamination was strikingly different. The counts on the aft table far exceeded all the others and cycled rapidly.

The results from experiment IX (Figure 12) show cyclical changes similar to those in experiment VIII, except in this experiment the highest level of contamination appeared on the floor of the personal hygiene area.

A composite graph (Figure 13) gives a somewhat more simplified representation and enables the visualization of the main trends. From this graph, it is seen that the basic trend is upward until day 25, with the most rapid and consistent rise occurring on the table. The counts from both the table and the floor of the personal hygiene area reached a maximum at the same time. This was not

unexpected, since the same air was circulating through all areas. The peak in bacterial contamination at day 25 presents interesting evidence of interaction between man and environment, since the maximum bacterial counts for both the groin and axillar areas peaked at the same period.

#### CORRELATION ANALYSIS BETWEEN STAPHYLOCOCCI AND CORYNEBACTERIA

An analysis to determine whether any correlation existed between staphylococci and corynebacteria was performed. This analysis depends on the assumptions that the treatment and environmental effects are additive, and that the experimental errors are independent in the probability sense, and are normally distributed. Correlation indicates the extent that the two microorganisms are related to each other. The correlation coefficient is a relative measure of the degree of association between two series and independent variables are always uncorrelated.

The groin, previously determined to be an excellent indicator area for bacterial buildup, was chosen to test the relationship between staphylococci and corynebacteria. This study involved the analysis of 283 separate pairs of values, using the formula

$$r = \frac{S_{XY}}{S_X S_Y}$$

where

$$S_{XY} = \frac{\sum_{i=1}^n X_i Y_i - N \bar{X} \bar{Y}}{N}$$

and

- $r$  = correlation coefficient
- $s_x$  = standard deviation of X
- $s_y$  = standard deviation of Y
- $\bar{X}$  = average of X
- $\bar{Y}$  = average of Y

The correlation coefficients fell within a range of  $r = 0.53$  to  $r = 0.35$  for each group, with an overall value of  $r = 0.43$ . Since perfect correlation occurs when  $r = +1.0$  or  $r = -1.0$  (and no correlation exists when  $r = 0$ ) the value 0.43 does not indicate any strong direct relationship, although it does indicate some minor degree of common association which can be accounted for by the common "treatment" exerted on both; namely, the lack of washing and the concurrent increase in numbers of bacteria.

Selected samples from the axilla were subjected to the same type of analysis, and the results showed the same minor degree of correlation.

The absence of significant statistical correlation does not preclude the existence of some definable relationship between these two organisms, since other more powerful tests may indicate such a relationship. To ascertain what relationship, if any, exists, a complete regression analysis is required. This can determine what proportion of the total variance is attributable to each of the variants. However, for the amount of data to be processed, the lengthy arithmetic calculations which this analysis requires could reasonably be attempted only with a computer program. Preliminary analysis on a limited number of samples indicates that this might be a worthwhile investigation.

Summarizing, the maximum information from the numbers of colonies counted over the 2-year period of the seven experiments, was obtained by calculating averages to indicate the number of bacteria present on a particular day. By using the average as a point of reference, the variability of the counts was determined, and the statistical significance of the variability was established.

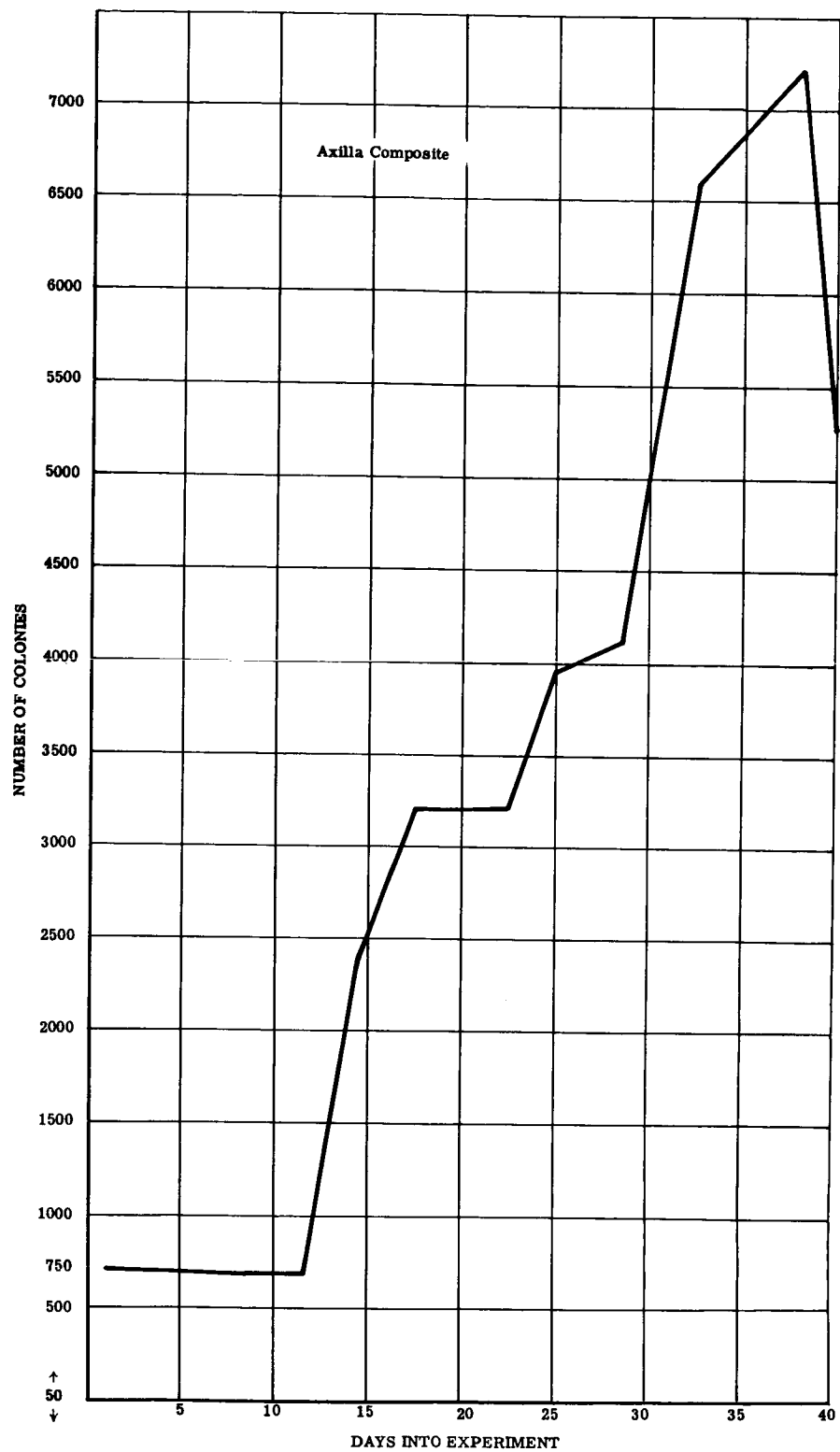


Figure 3. Axilla Composite



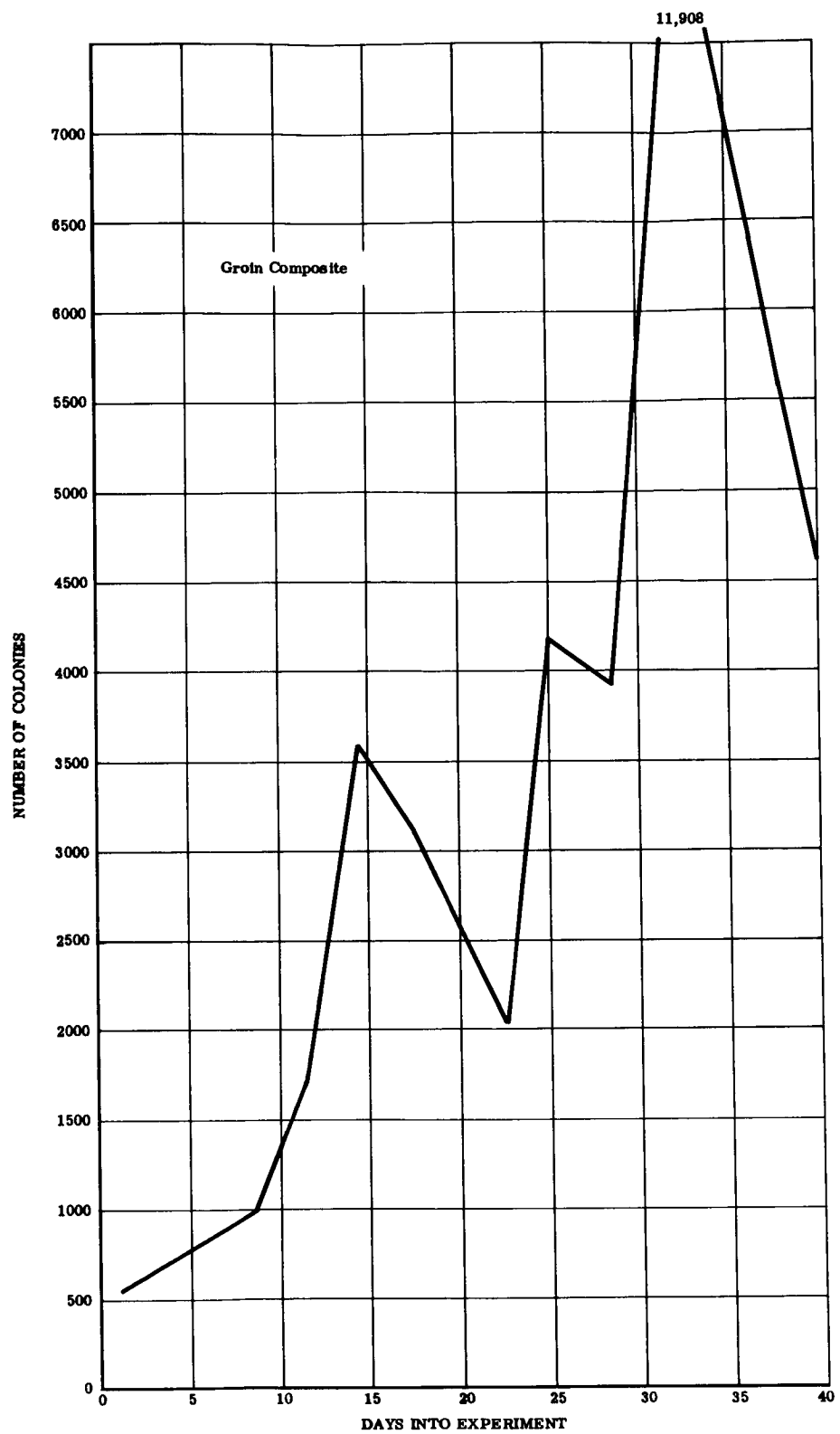


Figure 4. Groin Composite

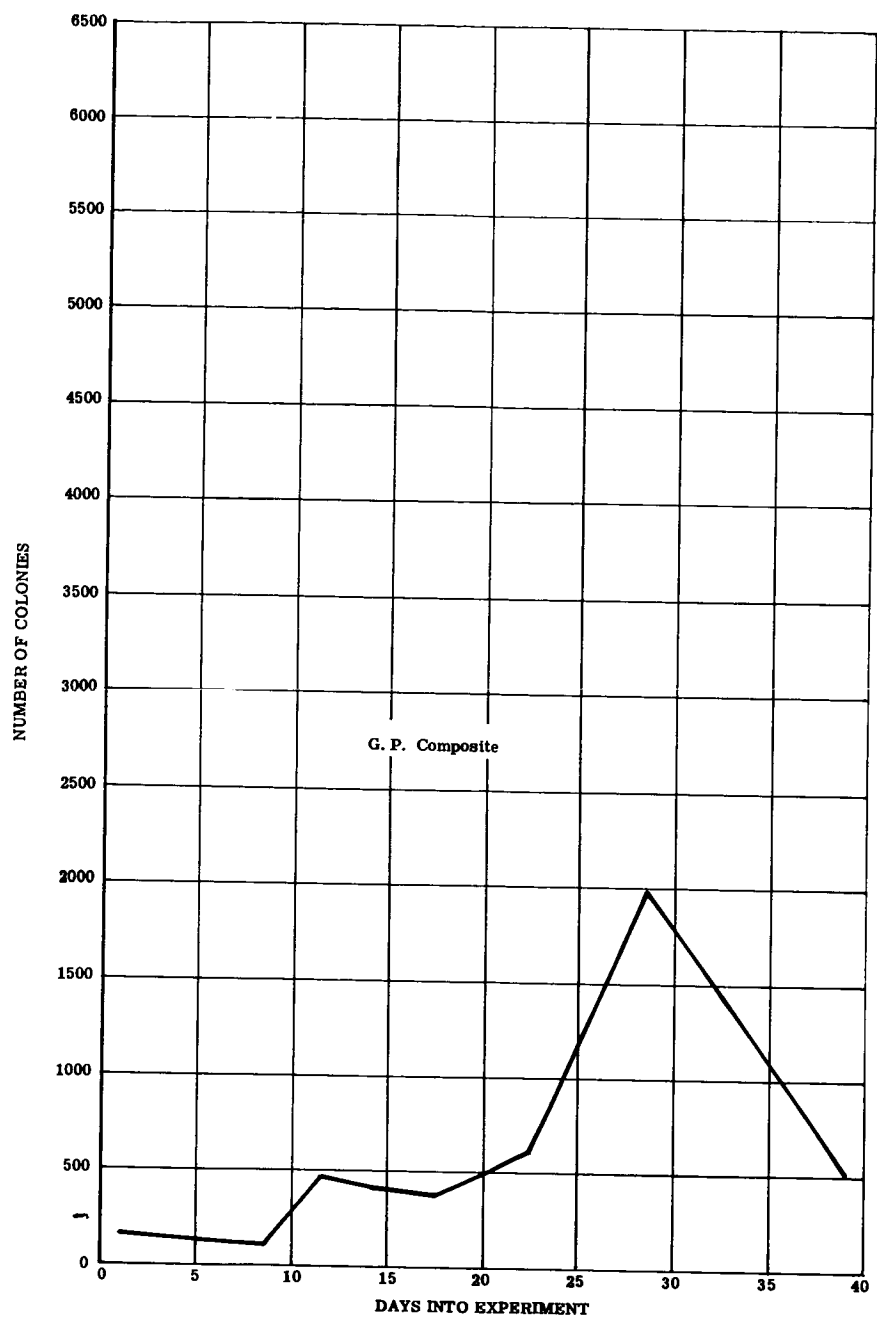


Figure 5. Glans Penis Composite

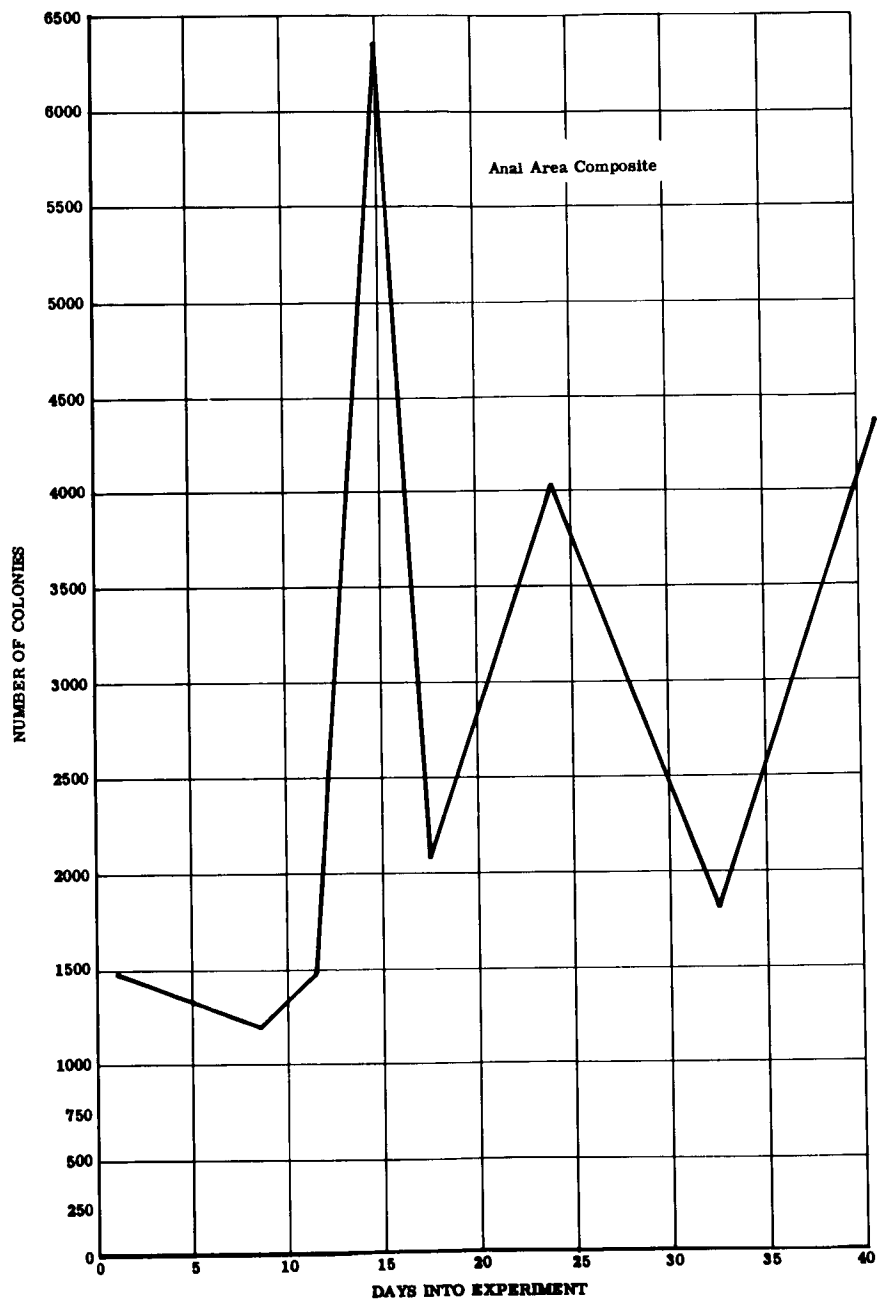


Figure 6. Anal Area Composite

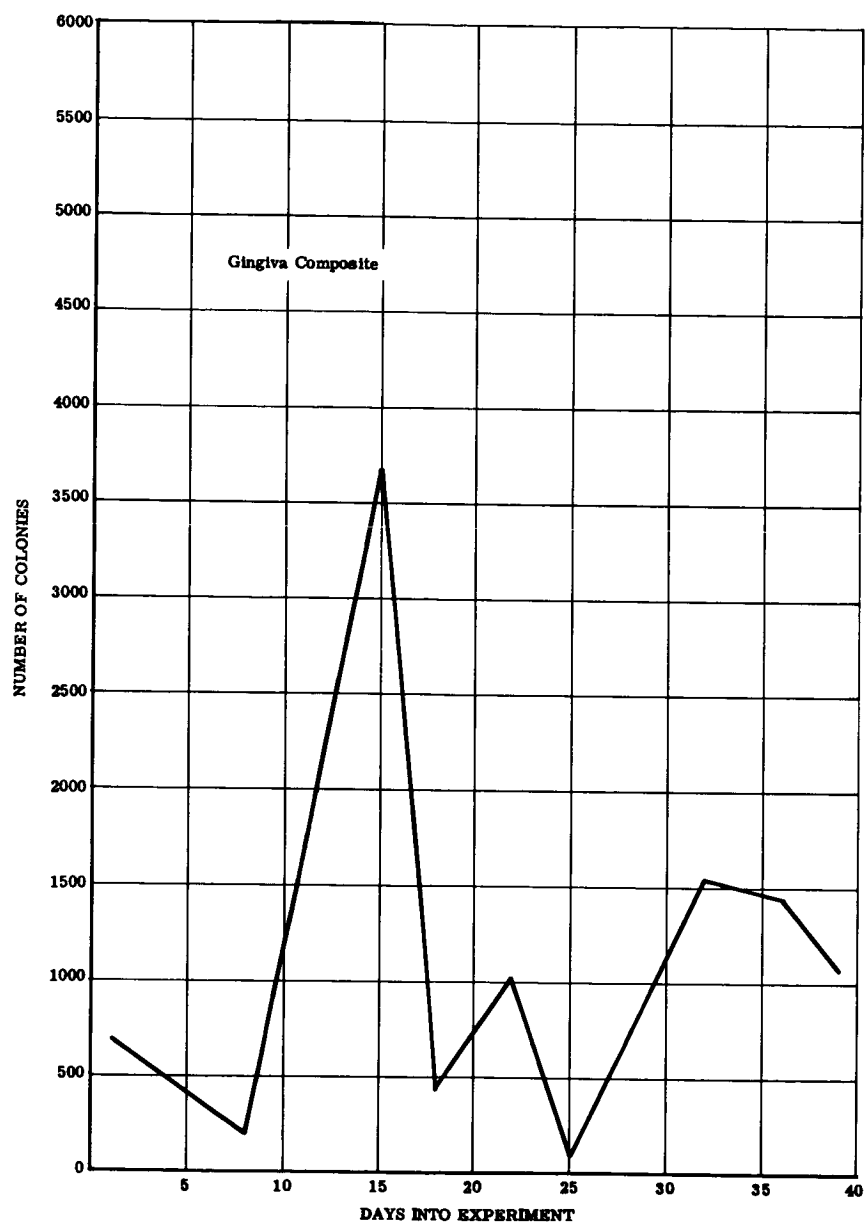


Figure 7. Gingiva Composite

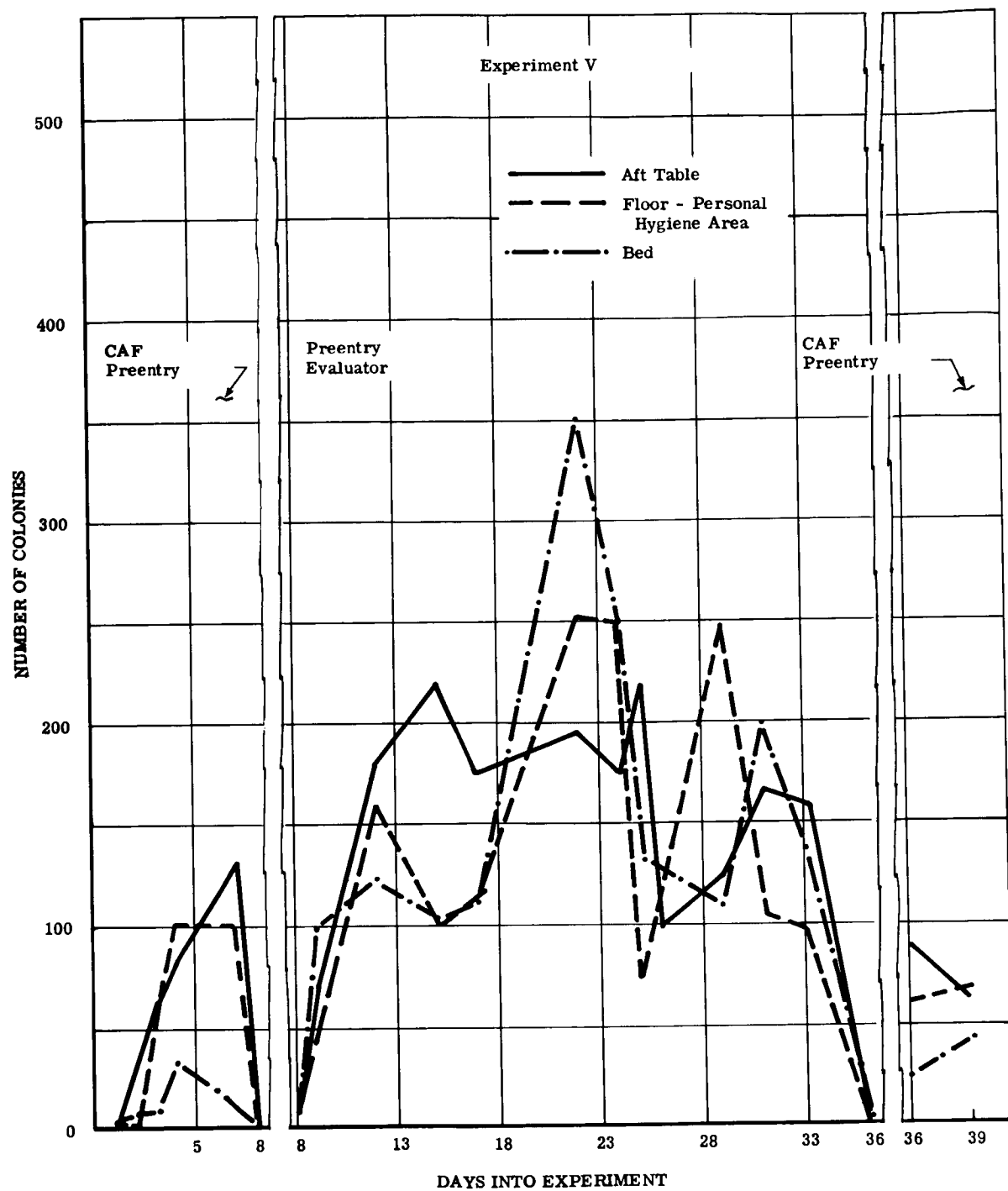


Figure 8. Experiment V - Environmental Areas

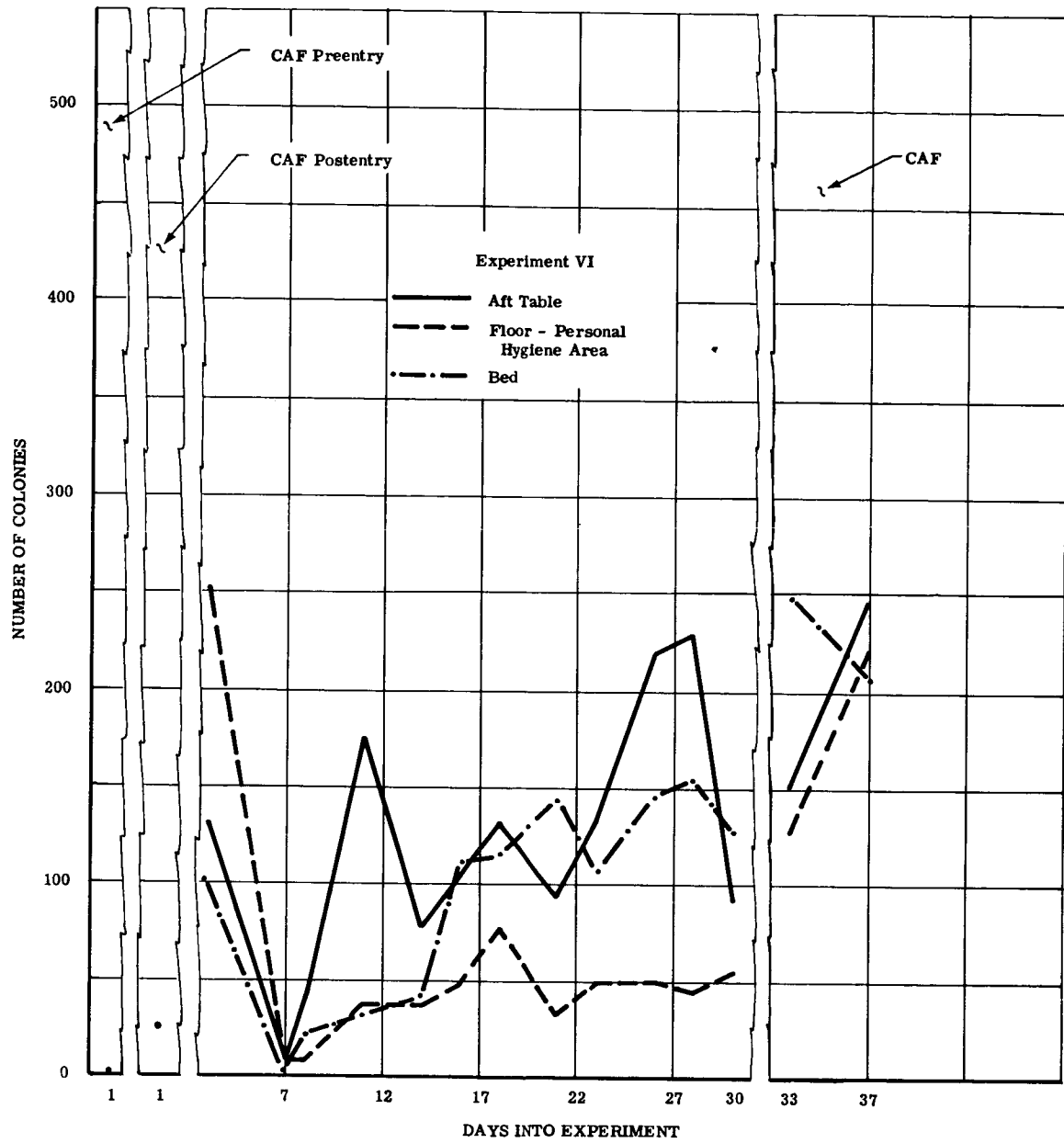


Figure 9. Experiment VI - Environmental Areas

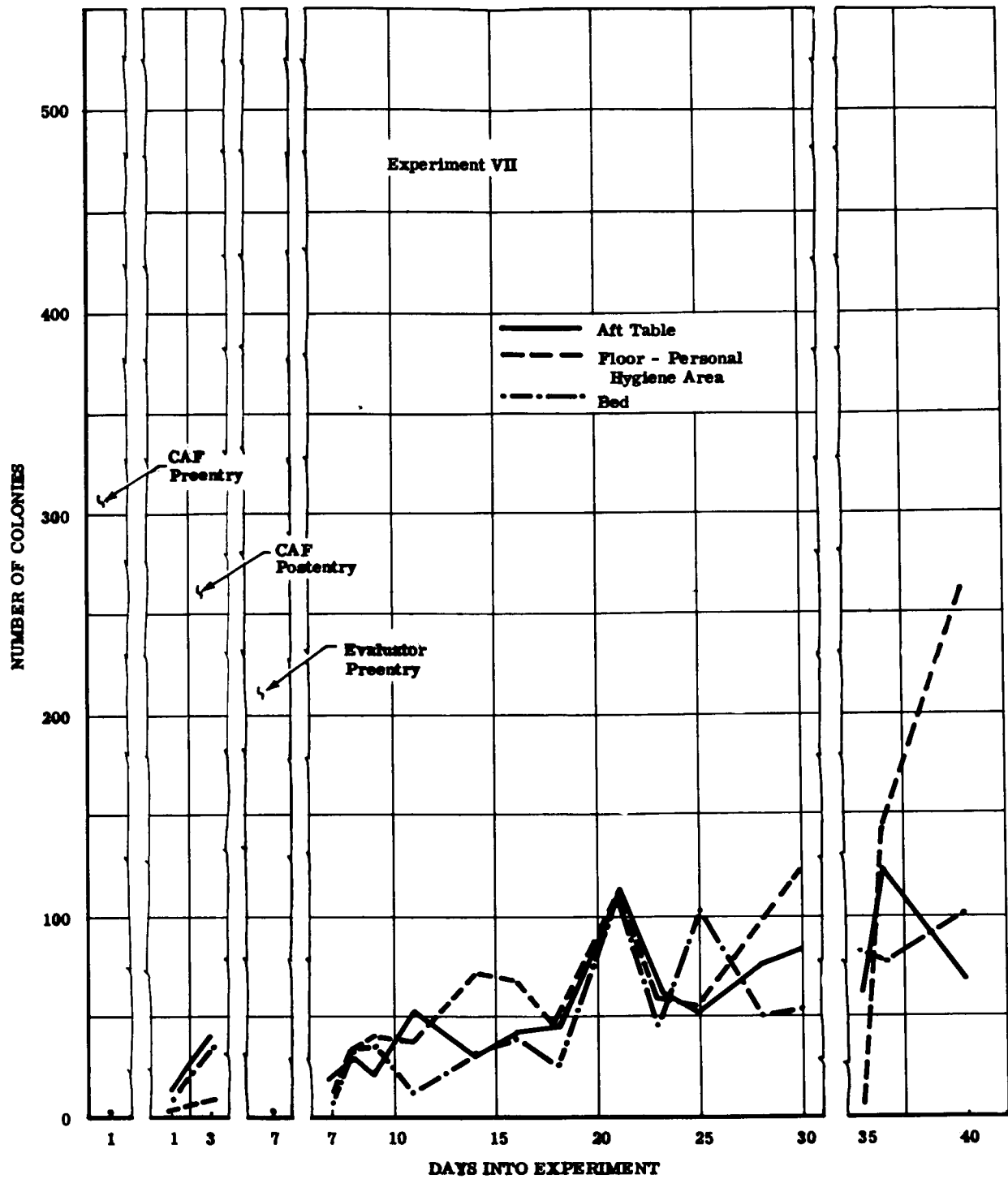


Figure 10. Experiment VII - Environmental Areas

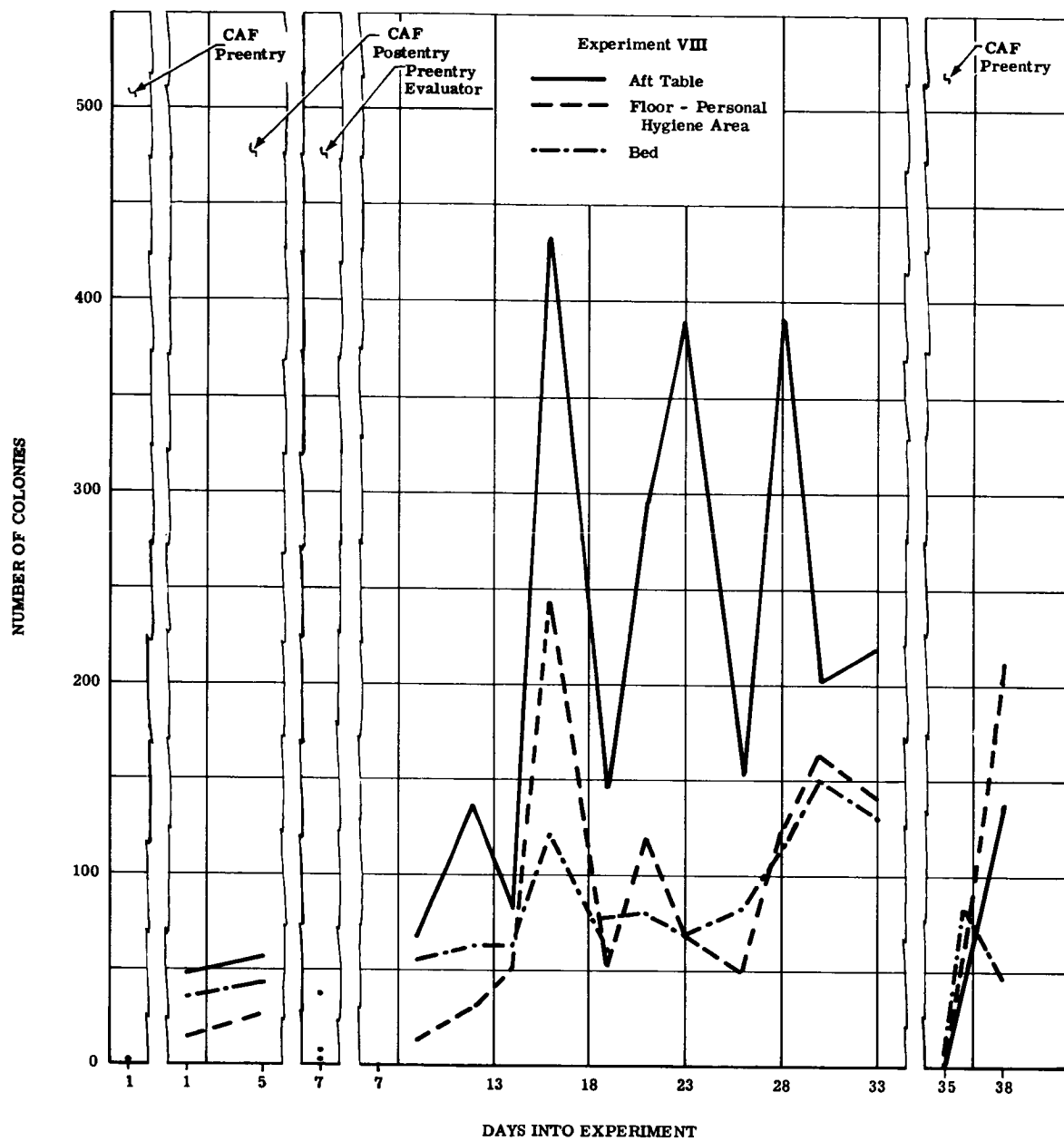


Figure 11. Experiment VIII - Environmental Areas



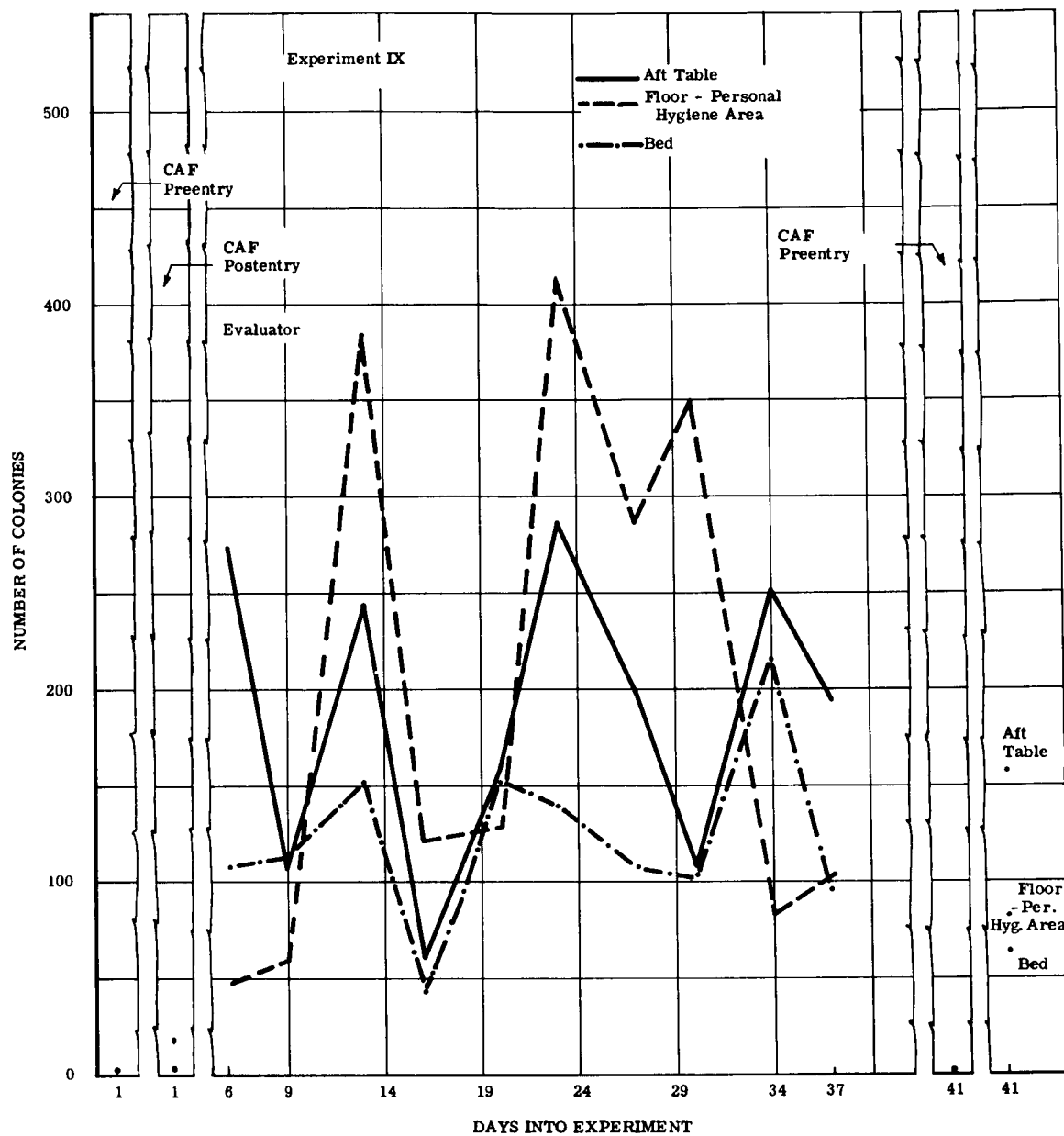


Figure 12. Experiment IX - Environmental Areas

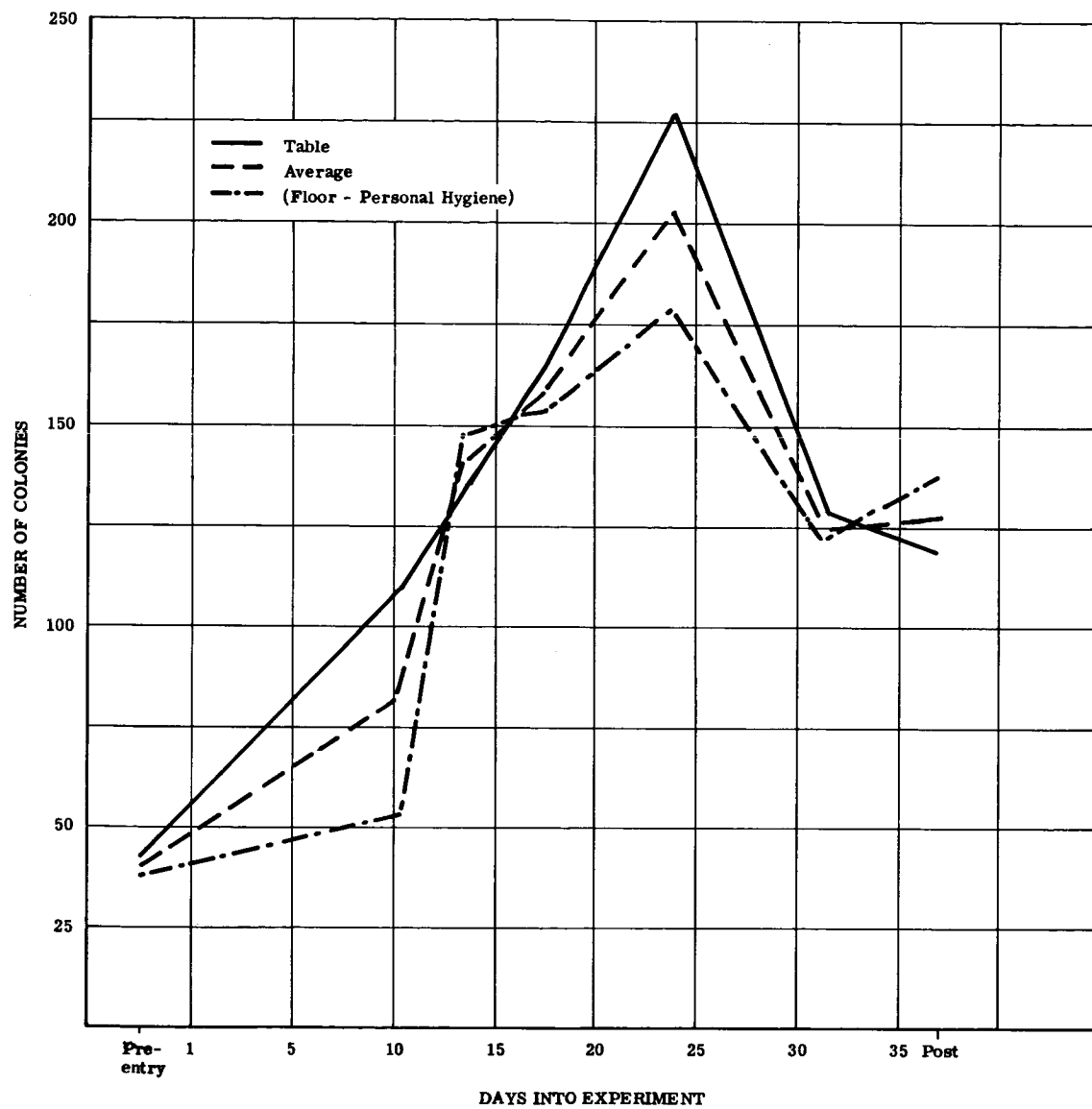


Figure 13. Composite Graph of Environmental Areas

## SECTION V

### EXAMINATION AND IDENTIFICATION OF NONSPORULATING FECAL ANAEROBES

The fecal flora is influential to the health and well-being of humans. The complex nature of this intestinal microflora, composed of more than 60 different species, contributes indirectly to the following functions: (1) host susceptibility to enteric infection<sup>(1)</sup>, (2) malabsorption of dietary fat<sup>(14)</sup>, (3) vitamin B<sub>12</sub> absorption or malabsorption<sup>(15)</sup>, (4) shock<sup>(16)</sup>, (5) hepatic coma<sup>(17)</sup>, (6) resistance to radiation<sup>(18)</sup>, as well as the ordinary processes of digestion.

During studies of fecal flora of men on defined diets the most drastic changes occurred, not in the numbers, but in the kinds of nonsporulating anaerobes predominating in the higher dilutions of fecal material. Because these changes could be caused by many factors, including the nature of the culturing schema, and the laboratory identification, the methods and techniques being used were carefully reviewed. An ideal technique for studying fecal flora would be one which could provide consistently valid information concerning the numbers, kinds, and physiological function of the fecal flora and one from which the information would be applicable to the in vivo rather than the in vitro situation.

Specimens were promptly obtained from the donor and cultures were made within 15 minutes, since, as Donaldson<sup>(15)</sup> has reported . . . "Unless specimens are properly diluted and cultured with specific mediums under appropriate conditions, nonsporulating anaerobes and lactobacilli will not grow even though these organisms may be present in large numbers. The rapid growth of coliform organisms on a variety of artificial mediums frequently obscures the presence of other slower growing species."

In previous Republic studies<sup>(5, 8, 12, 13)</sup> anaerobic cultures were assigned to groups by morphological and biochemical characteristics and designated by FA or GD numbers. The necessity for this approach was based upon the lack of pertinent schema which would allow these organisms to be readily identified. In addition, this approach made it possible to screen large numbers of obligate

anaerobes, which could then be assigned to these arbitrary groups. The ability to handle large numbers of cultures facilitated the screening for shifts in predominating groups of nonsporulating anaerobes. In addition to observing the shifts in fecal populations, it allowed rough correlations of anaerobic shifts with dietary changes. The dietary changes were either in format, composition, or total amount. The physiological implication of these changes in fecal flora has not been well defined, and much work must be done both in vivo and in vitro to assess the detrimental or beneficial effect of such changes.

The space-type diets used in nutritional experiments were correlated with shifts in the predominating fecal anaerobes and as stated by Vanderveen et al.,<sup>(19)</sup> "Observations made on the effects of the diet on the gastrointestinal tract of the crew members indicated the diet had a low compatibility for space use. The data ... show that each subject had an average of one fecal specimen for each day on the diet. The majority of specimens were nonformed, had a pungent odor, and reportedly caused difficulty with personal hygiene. Note the unusually low water content of the fecal matter considering that most stools were nonformed. The fat level in the specimens was unusually high for a diet of a moderate fat intake. Clinical tests for indications of malabsorption of fat, such as urinary indicans, were normal. During the low pressure phase of the experiment, the crew members reported problems with flatus production. Upon several occasions, distention caused by gas in the gastrointestinal tract became so severe that the crew members could not perform in an efficient manner." Since the subjects in this study lived in an oxygen-helium atmosphere with differing atmospheric pressures, this factor may partially explain the difficulties they encountered. However, in a paper by Slonim<sup>(20)</sup>, reference is made to a statistically significant increase in fecal fat in men on compressed bite-sized foods, and the description of the fecal specimens agrees with those described by Vanderveen. Since the study by Slonim is based on the same experimental data as this study, it is interesting to speculate whether the microbial changes observed in the fecal material of men on this diet were a result of, or were responsible for this low diet compatibility.

The numbers, kinds, and changes in these predominating anaerobes are well documented<sup>(13)</sup>. In efforts to interpret the possible medical significance of these changes, it was considered essential to identify these anaerobes by recognized

classifications. Identification into a recognized classification sounds very simple; however, it is exceedingly difficult. For example, recognized authorities in the field differ widely in the classification of nonsporulating anaerobes. A. Trevor Willis<sup>(21)</sup> states that all anaerobic gram-negative nonsporulating bacilli should be included in the genus *Fusiformis*. This is in direct contradiction to Bergey's Manual<sup>(6)</sup> which divides the gram-negative anaerobes into *Bacteroides* (those with rounded ends) and *Fusobacteria* (those with pointed ends). To further complicate the picture, it is necessary to realize that "fusiform," which is a morphological description, does not necessarily indicate that the organism in question belongs within the recognized *Fusobacterium* classification<sup>(22, 23, 24, 25, 26, 27)</sup>. To add to the confusion, Rosebury<sup>(28)</sup> places all nonpleomorphic nonmotile nonsporulating (saccharolytic) anaerobes into the species *Bacteroides fragilis*. These systems of classification are comparatively simple to that of Prevot<sup>(29)</sup> who has divided these organisms into several hundred species, often on the basis of a single isolation and limited biochemical identification. The works of Prevot and Bergey and other authors<sup>(30, 31)</sup> were used as the basis for keying of the predominating fecal anaerobes done in the study reported herein. As shown in Table 31, an expanded biochemical schema for identification was used. Representative cultures of the FA and GD types were classified according to this schema, as were cultures obtained from the American Type Culture Collection (Table 32). Cultures representing certain genera were not available from the American Type Culture Collection, and could not be included.

A basic step in the identification of the predominating fecal microflora is the dilution series. These series are either aerobic or anaerobic, depending upon the media and method of incubation, and are carried out in the manner detailed by Gall et al<sup>(8)</sup>.

The importance of the dilution series in the isolation of predominating fecal anaerobes is well shown in Figure 14 which includes many photographs of incubated anaerobic cultures from a dilution series of two subjects who were confined and who were on a defined space-type diet (Table 1). The caption under each photograph is a key. The first number indicates the subject's code number, the dash number following the word "Feces" is the number of the fecal specimen, while the number in parentheses indicates the dilution of the sample. The four different fecal samples

of subject 41 show the changes in predominating fecal anaerobes corresponding to the length of time he was on a particular diet. The variation of the bacterial population between the two subjects may be noted by comparing the photographs for subject 41 with those for subject 44 at the ninth sampling period. By the 16th sampling period, the bacterial populations became more complex. In addition to the original anaerobes, several other new types appeared.

Symbiotic relationships are apparent in the morphological character of the bacteria. When these bacteria are isolated in pure culture, the individual morphology often varies and is less distinct, probably because the researcher is unable to supply the complex nutrients essential for each species.

Before identifying any organism, it is necessary to ensure that the culture in question is, in reality, pure. It must be free of both facultative aerobic and anaerobic contaminants. Methods of purifying anaerobic cultures are dependent upon the laboratory in which the study is being conducted. In this laboratory, a pour plate method was found to be the most satisfactory. In this method, a thin layer of Gall's anaerobic agar is poured into a plate, a broth dilution series of a culture (thought to be mixed) is made, and 0.1 ml of the dilution is placed on the hardened anaerobic layer. An additional layer of Gall's anaerobic agar is poured over the overlay. The plates are placed in an anaerobic jar, which is evacuated, flushed with 10% CO<sub>2</sub>, re-evacuated, flushed with H<sub>2</sub>, N, CO<sub>2</sub> mixture, then incubated at 37C. In addition, conventional Brewer pour plates are made from the dilution series. In some instances, when cultures were extremely difficult to purify, a sterile glass tube was filled with Gall's agar in which a minute portion of inoculum had been placed. Following incubation at 37C in a CO<sub>2</sub> incubator, the contents of the tube were expelled into a sterile petri dish and discrete colonies were easily isolated. In addition, control aerobic plates from all cultures were inoculated and incubated aerobically. Certain microaerophilic organisms produce small surface colonies under aerobic conditions, and certain microorganisms are obligately anaerobic only on primary isolation, and become oxygen-tolerant after two or three subcultures. Therefore, replication of each procedure must be performed before a valid conclusion can be reached. Selectivity was used in determining which procedures should be included in the differential schema.

Cellular morphology was recorded from all cultures at various times during the incubation period. Many of these anaerobic species are extremely pleomorphic and forms varying from coccoid to long filamentous rods are present in a particular species. For this reason, phase variation is an important consideration in describing microscopic morphology.

Following purification, the various tests and methods which would provide the most useful information for classifying the microorganism were used. The Gram reaction was not stressed, since it is of little importance in describing anaerobic cultures, as hourly variations are noted in the ability of these bacteria to retain the Gram stain. Of marked importance is the determination of spores, since the sporulating obligate anaerobes have been well studied and classified. Capsular and flagellar staining are also of little practical use in the routine identification of these anaerobes.

The absence of motility was not a key characteristic, since nonmotile variants of motile species often occur, and motility seems to be readily lost in culture. In addition, many of the delicate anaerobes refuse to grow in semisolid agar. Hanging-drop or wet-mount preparations are inadequate because of the oxygen effect.

Colonial, macroscopic morphology seems to vary even within subcultures from the same culture, and the size and shape of colonies will change depending on the period of incubation, the number of organisms involved, the moisture present in the media, and the concentration of agar.

Litmus milk was found to be an excellent medium for differentiating the various genera. The various changes produced in litmus milk by the nonsporulating anaerobes include acid, gas, a rapid curd formation, a slow curd formation, and subsequent digestion. In some organisms, a stormy curd is significant and easily recognized; this curd is produced by the rapid bacterial utilization of the lactose, with marked gas formation, disrupting the curd with gas bubbles. A curdling effect is not necessarily indicative of acid production, since some non-lactose fermenting organism secrete a rennin-like enzyme which hydrolyzes the casein to soluble caseinogen, which then reacts with the soluble calcium salts present in the milk to form a precipitate of calcium caseinogenate.

Many organisms, in their metabolism of proteins or protein-digestion products (cysteine taurine and other sulfur compounds) produce free  $H_2S$  in varying amounts. The  $H_2S$  can be readily detected in the medium by various methods. One very sensitive procedure used in this laboratory involves the addition of 0.1 cc of bismuth citrate to the medium. If  $H_2S$  is present, the ensuing reaction will result in the formation of bismuth sulfide, which is evidenced by a blackening of the medium.

Another key test used to separate the various genera involved the fermentation of glucose, lactose, maltose, sucrose, and dextrin. Two different sugar solutions were used to determine the pH. One is a 0.1% glucose heavily buffered; the other is a 0.5% glucose solution not buffered.) These sugars were used because they are characteristic of sugars present in the human digestive tract that are readily available to the microorganisms.

The anaerobic cultures were tested for their ability to reduce nitrate to nitrite. This test, as well as gelatin liquefaction growth on meat infusion agar, peptone water, serum dependence, fatty acid<sup>(31)</sup>, and indole production, was performed to compare results with those found in the literature.

Physiological characteristics of the FA type cultures determined in a previous study by this laboratory<sup>(12)</sup> are shown in Table 33. The deaminating, decarboxylating and lactic acid production ability of these anaerobes are important characteristics.

The results of all these tests were tabulated (Table 31). In addition, a table based on the findings reported in the literature was compiled (Table 34). Based on the biochemical reactions, the data from the literature, and the morphological characteristics shown in Figures 15, 16, and 17, generic names were assigned to the FA and GD series as shown in Table 35.

As anticipated by Gall et al.,<sup>(5)</sup> many of the fecal anaerobes fell into the same genera, and at times it was found easier to classify certain of the FA types into species. This was done wherever possible.

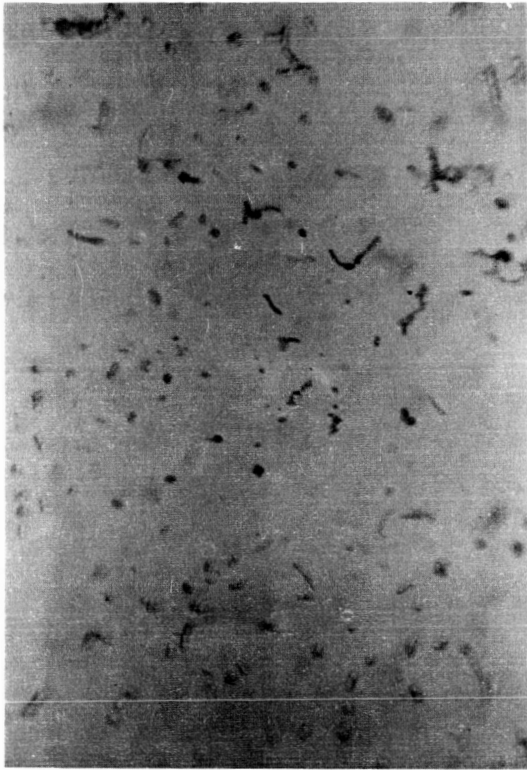
As shown in Table 35, only four of the FA/GD series fell into the genus *Bacteroides*: FA-7, FA-15, GD-3, and GD-6, while FA-3, FA-18, GD-1, GD-2,



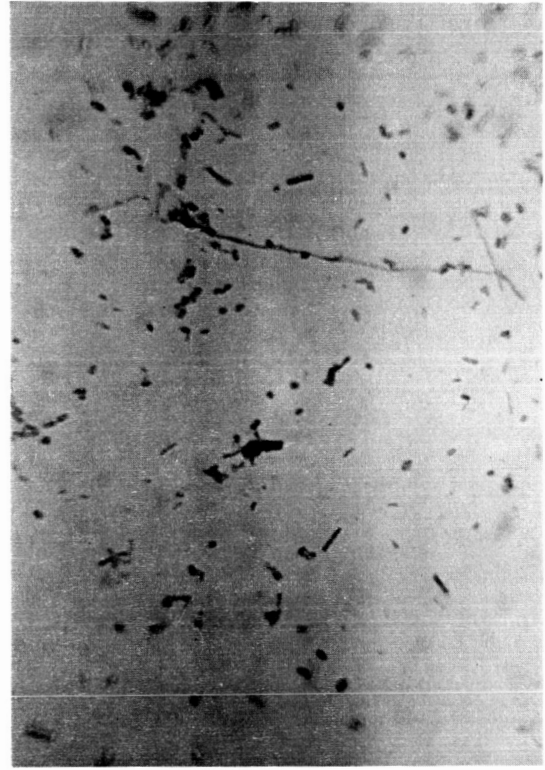
and GD-7 seemed to fit closely into the genus *Fusiformis*. *Sphaerophorus* is represented by FA-2, FA-10, FA-16, and GD-4. *Eubacterium* includes FA-4, FA-6, FA-11, and FA-12; FA-1 and GD-5 fall into *Catenabacterium*, while FA-9 and FA-17 appear in the *Ramibacterium* group. Four of the FA types represent different groups; FA-8, *Dialister*; FA-13, *Veillonella*; FA-4, *Butyri-**bacterium* (possibly *B. rettgeri*); and FA-5, *Lactobacillus*.

The identification of FA-2 as *Sphaerophorus* is based on a comparison of its biochemical reactions and particularly its morphology to that of the American Type Culture Collection culture of *Sphaerophorus* as studied in our laboratory. The characteristics differ somewhat from older, classical descriptions, but since there is agreement in biochemical determinations between our cultures and those supplied by American Type Culture Collection we feel this delineation of FA-2 as *Sphaerophorus* is justified.

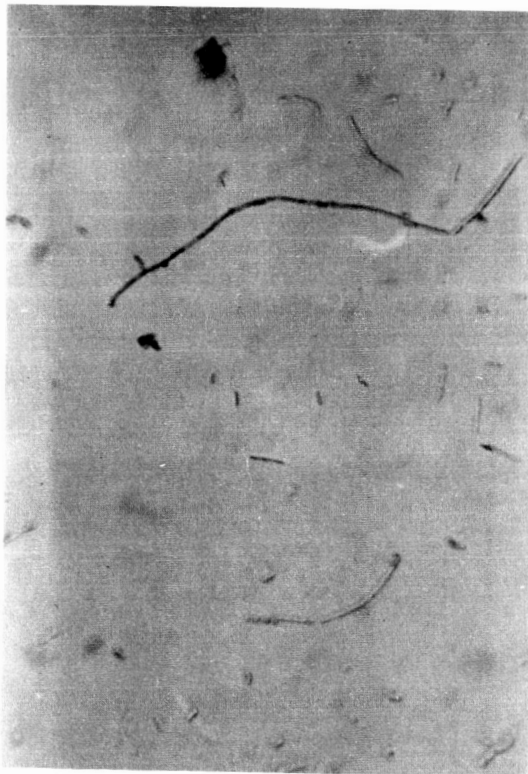
As this is going to print some basic taxonomic divisions within and among the *Lactobacillaceae* and *Propionibacteriaceae* are being questioned by the subcommittee on *Lactobacillaceae* of the American Society For Microbiology. Since their work is still in progress and no conclusions have been drawn it has not been used in designated generic classifications in this report.



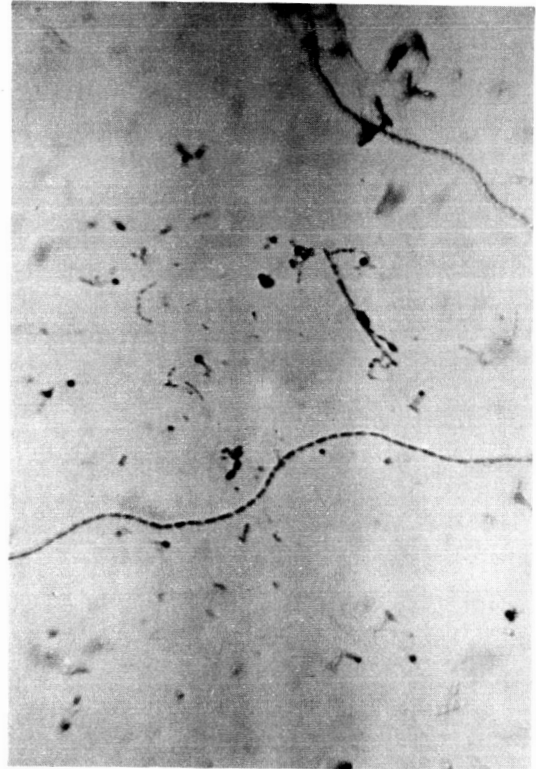
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41-Feces-6(4)



41-Feces-6(5)

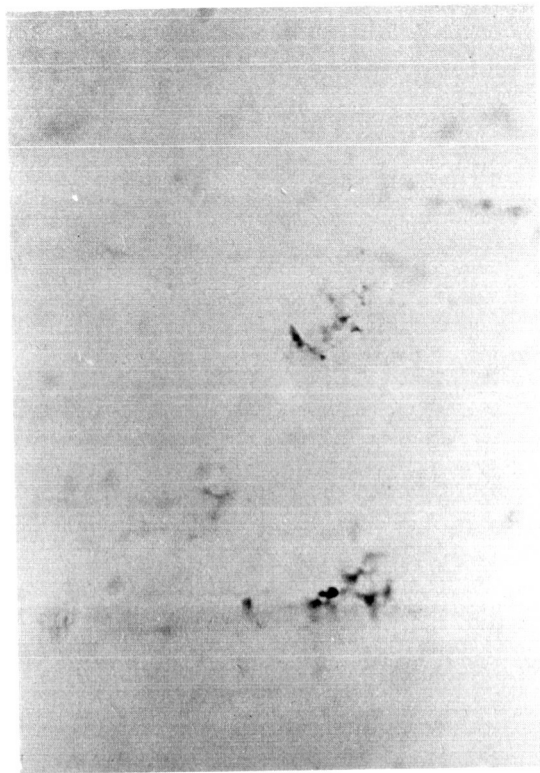


41-Feces-6(6)

Figure 14. Anaerobic Fecal Series

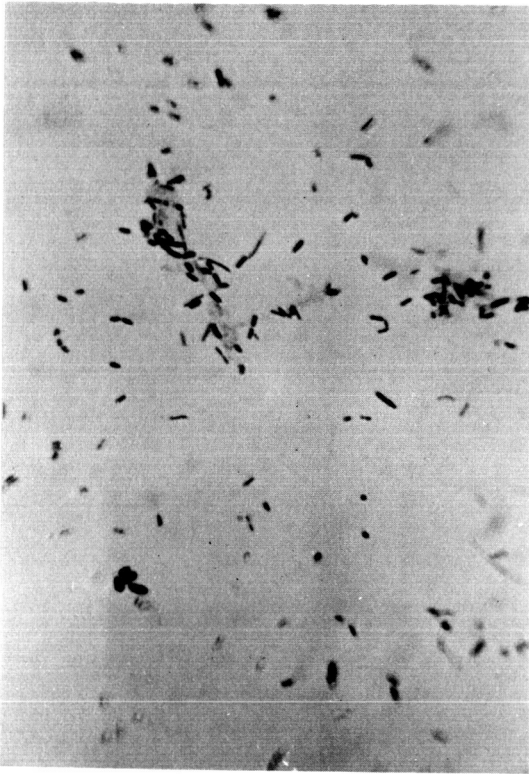


41-Feces-6(7)



41-Feces-6(8)

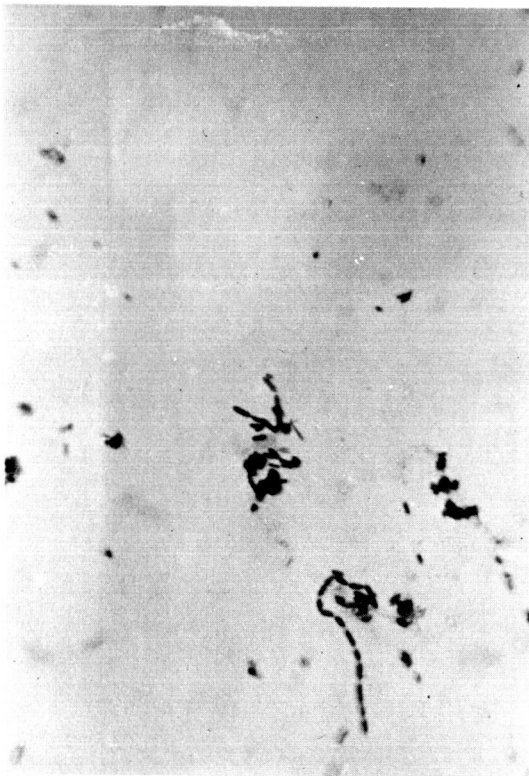
Figure 14 --- Continued



41-Feces-9(3)



41-Feces-9(4)

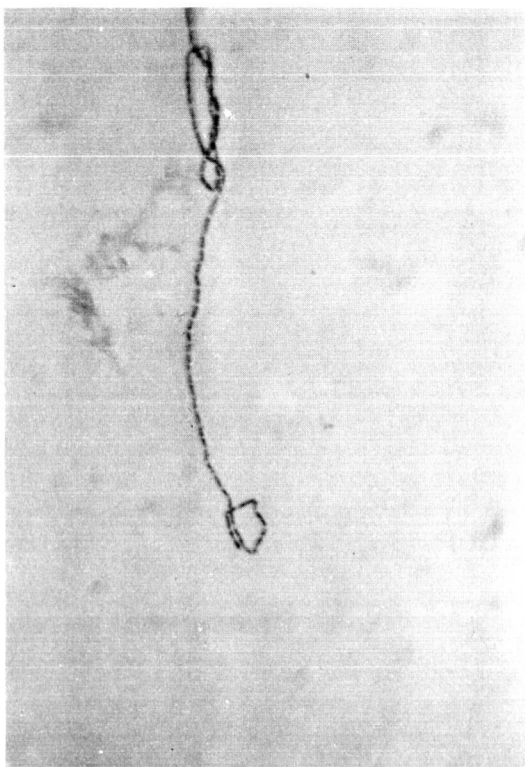


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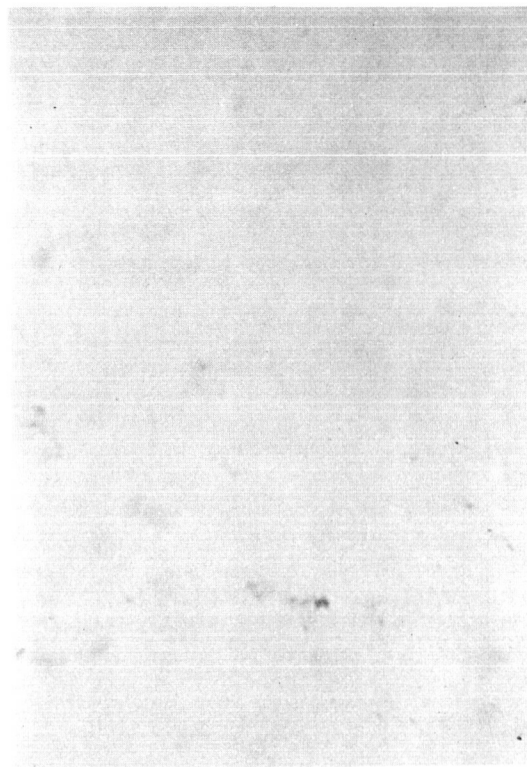


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Figure 14 --- Continued



41-Feces-9(7)



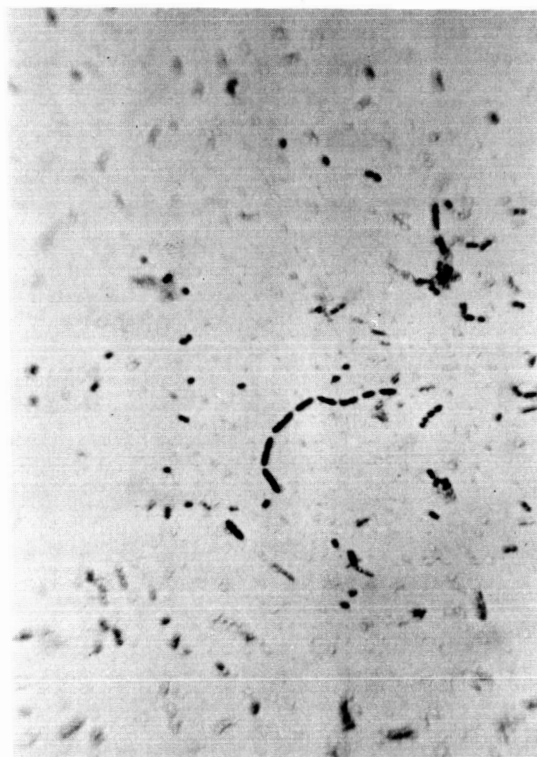
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Figure 14 --- Continued

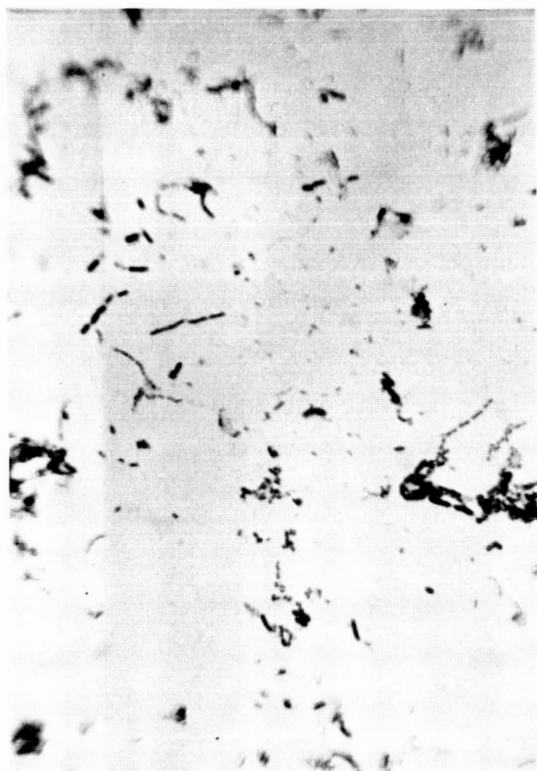




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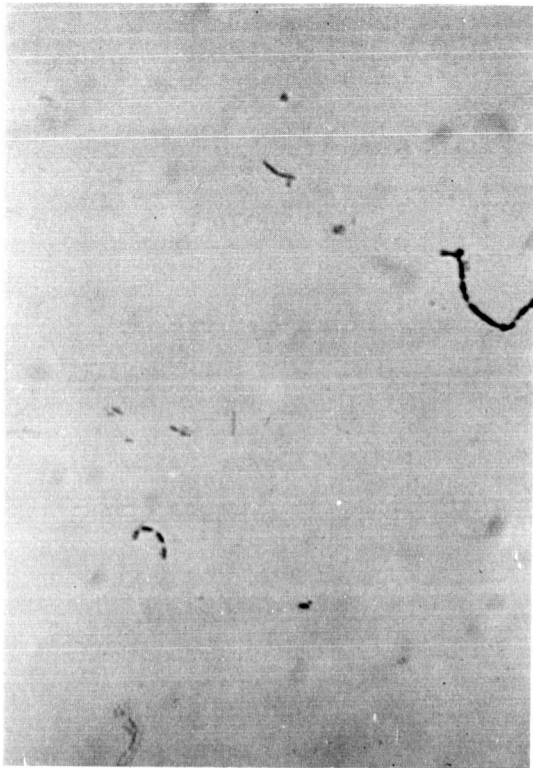


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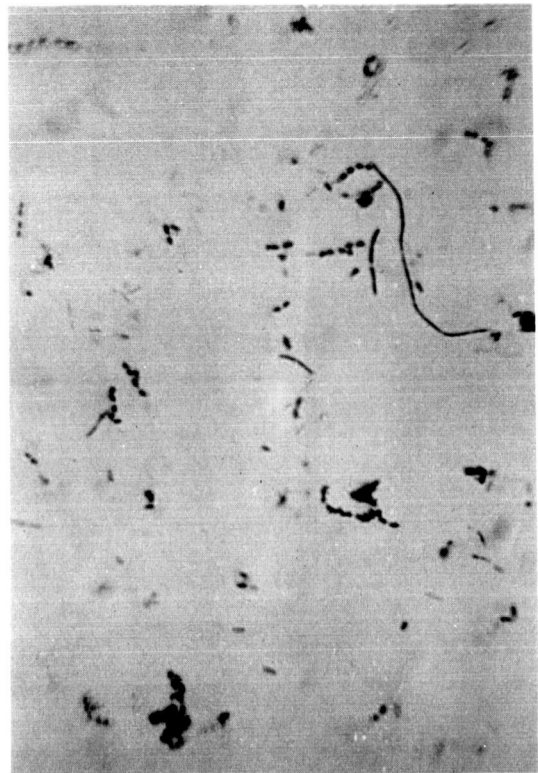


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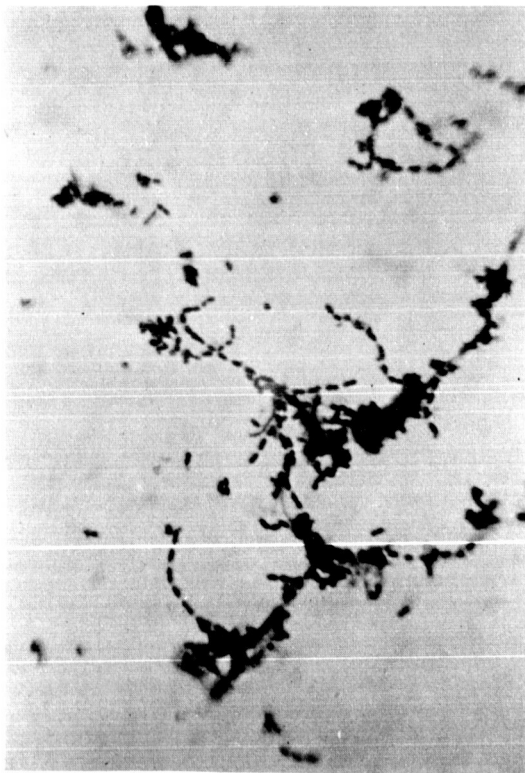
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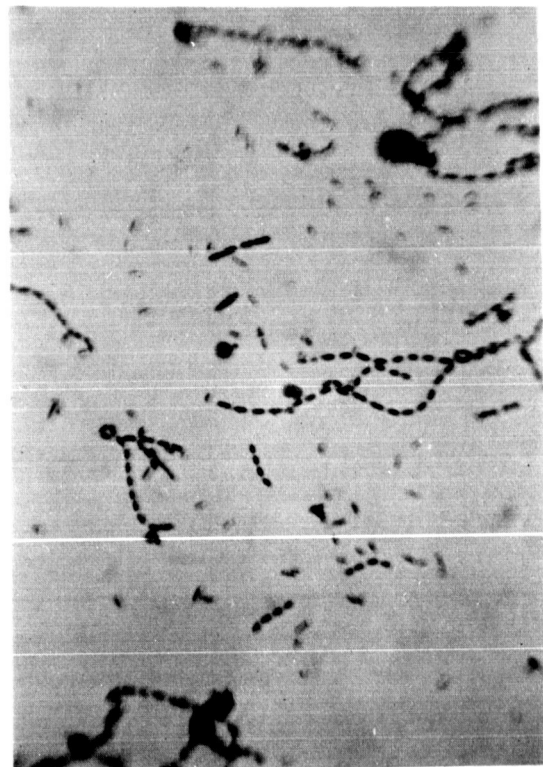
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44-Feces-4(4)

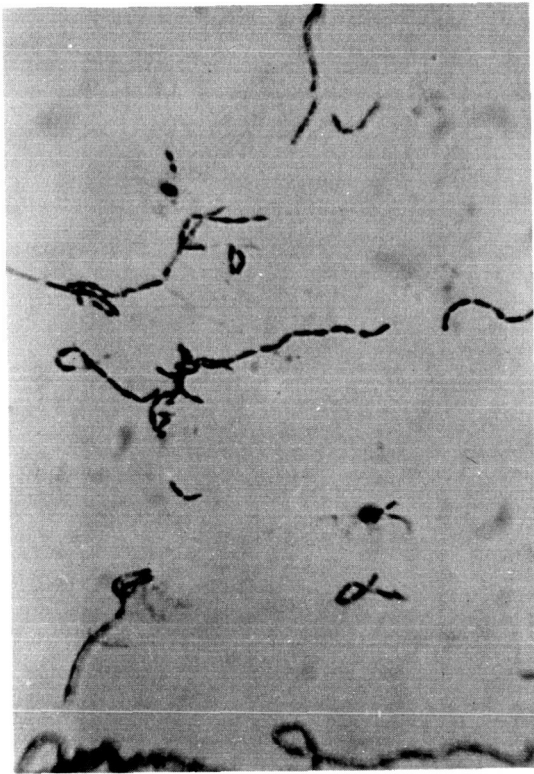


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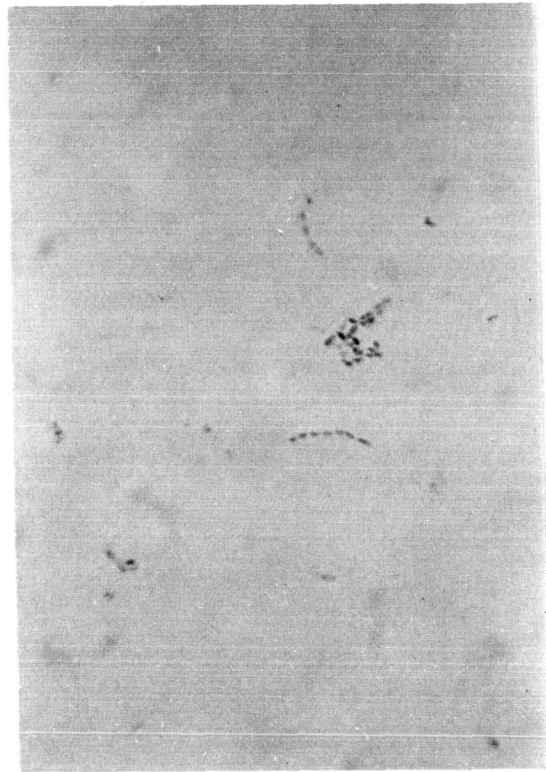


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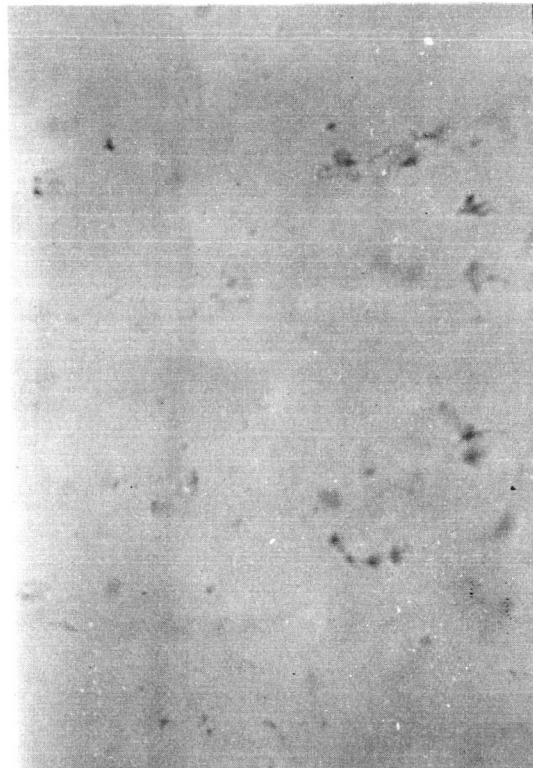
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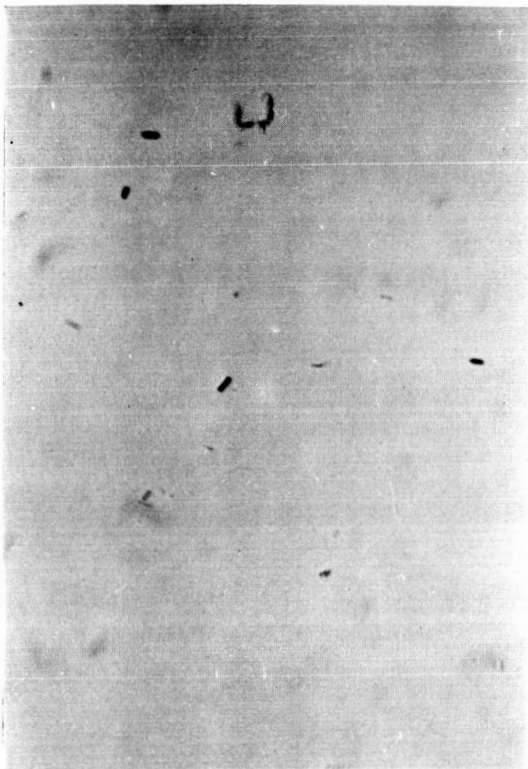
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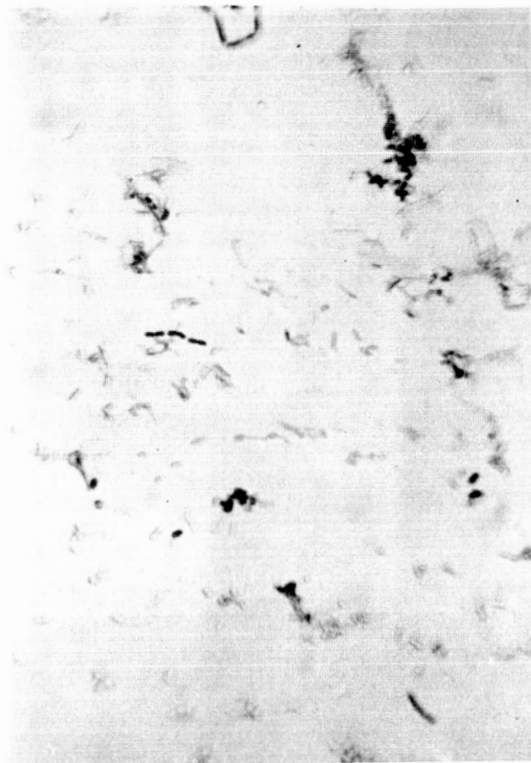
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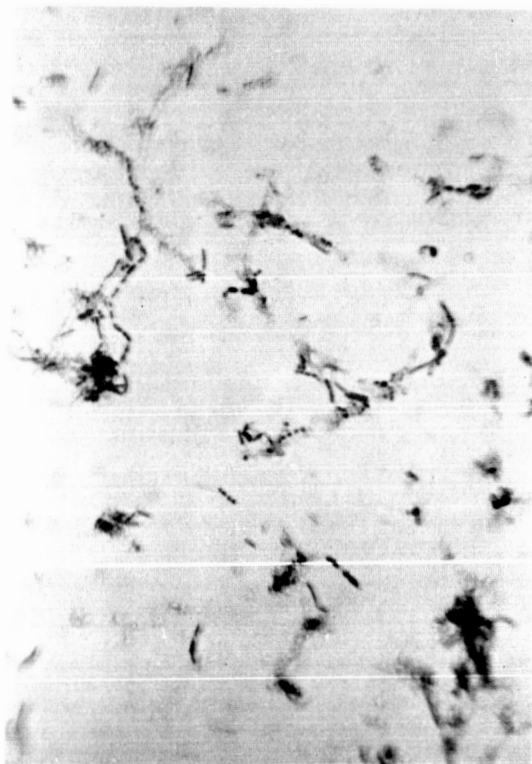




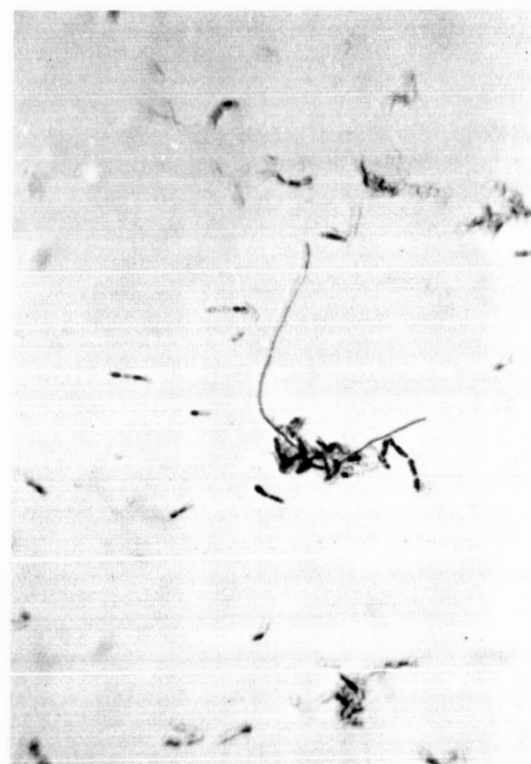
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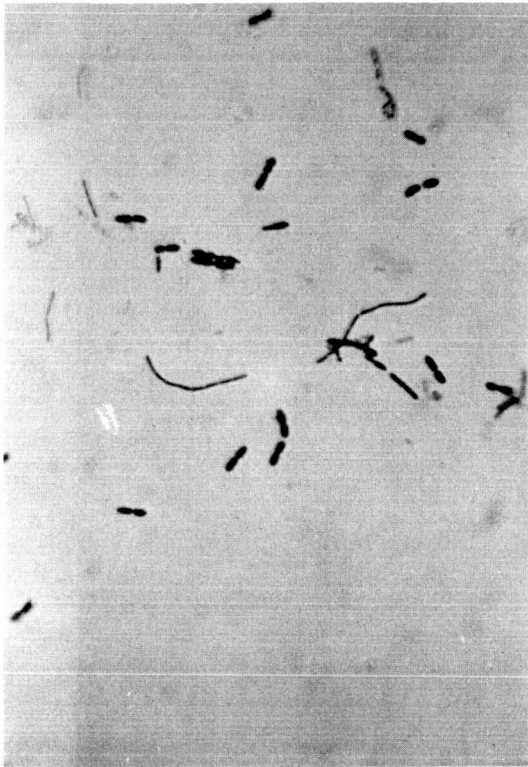


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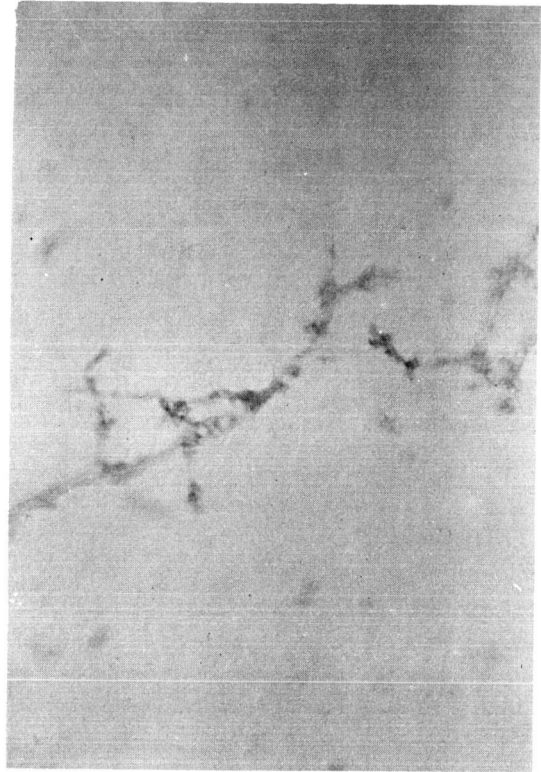


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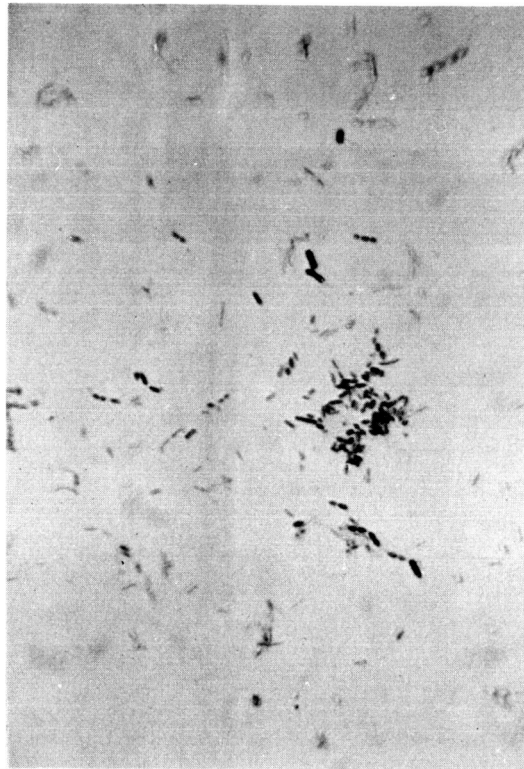
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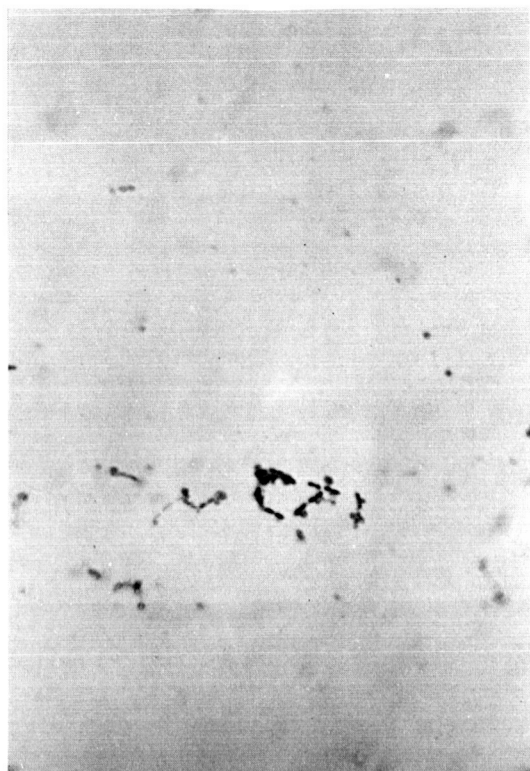


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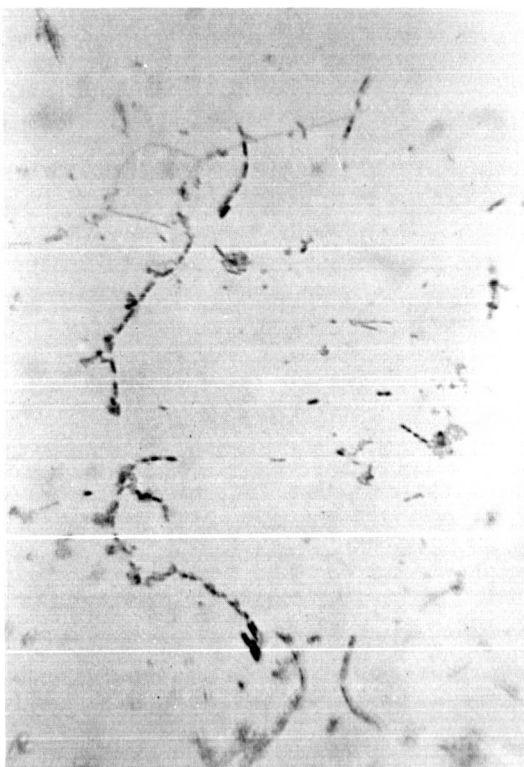


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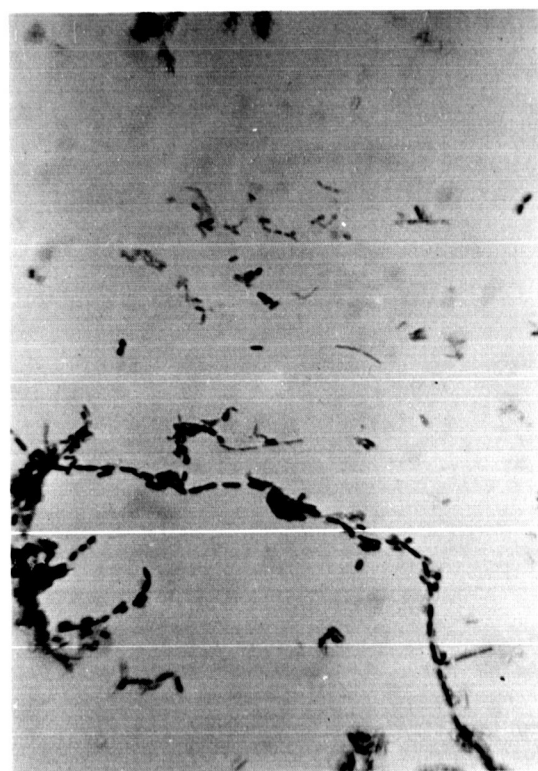
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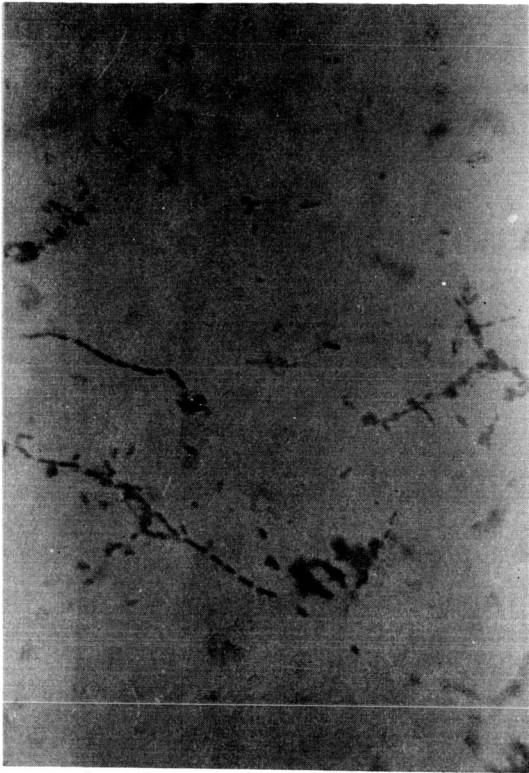


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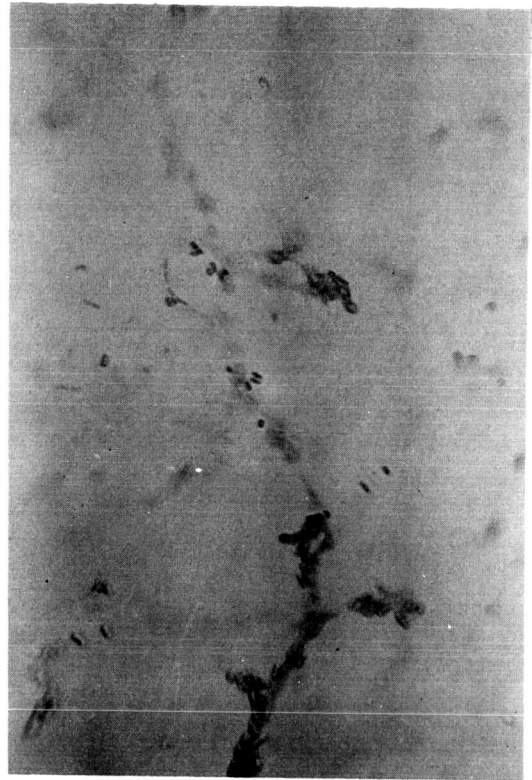


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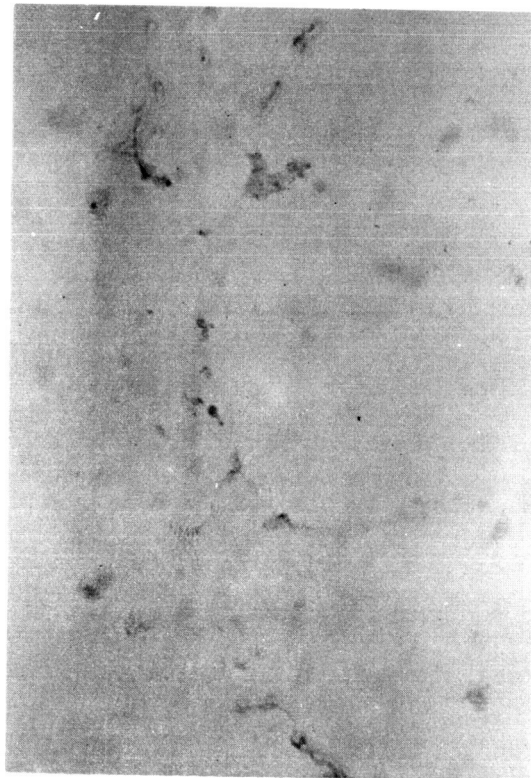
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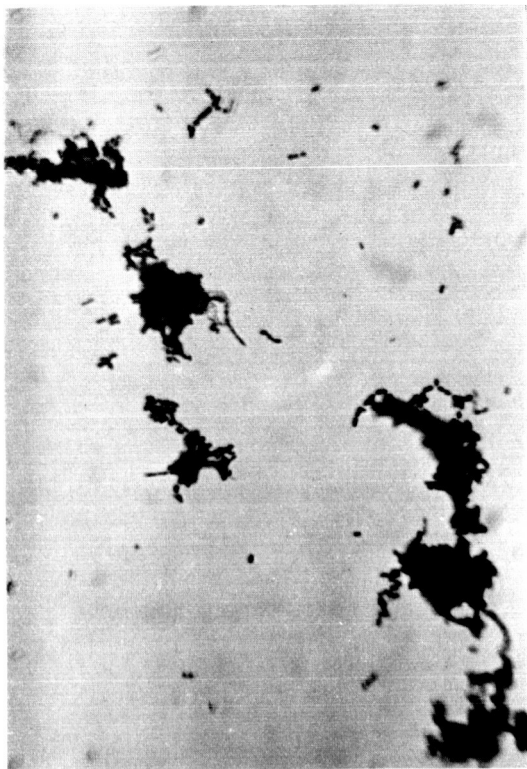
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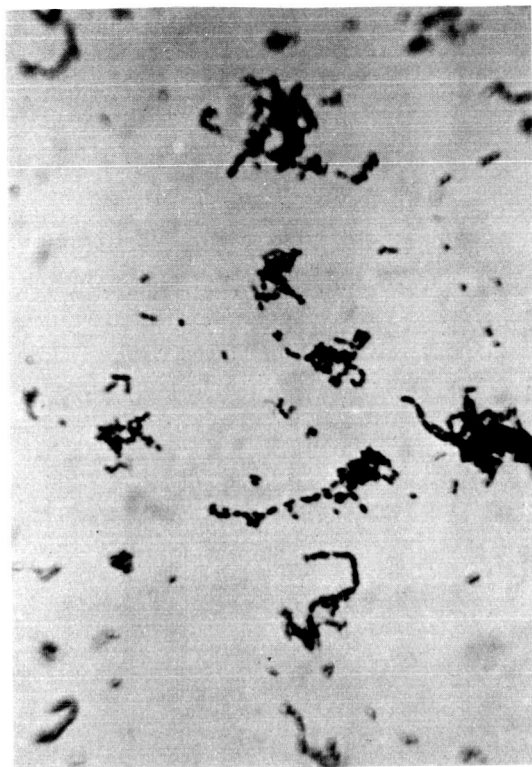
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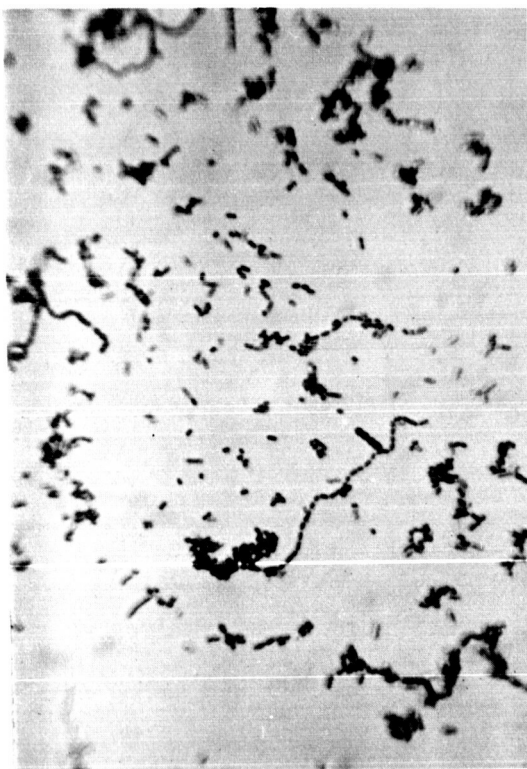




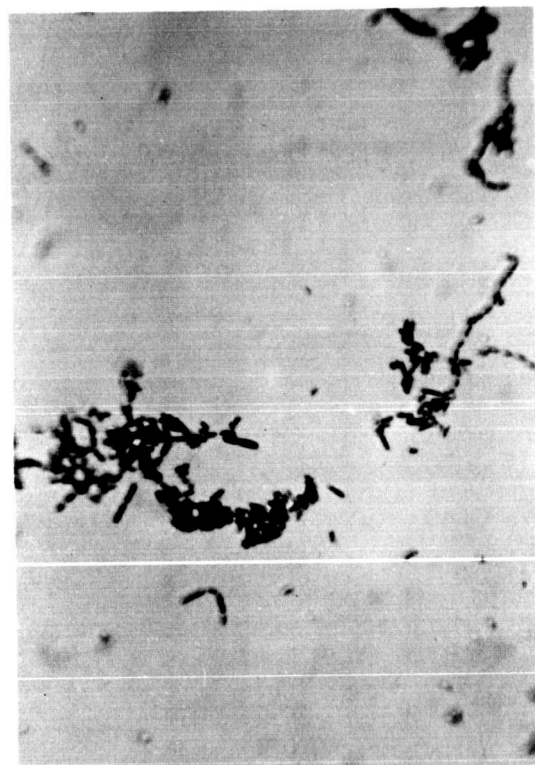
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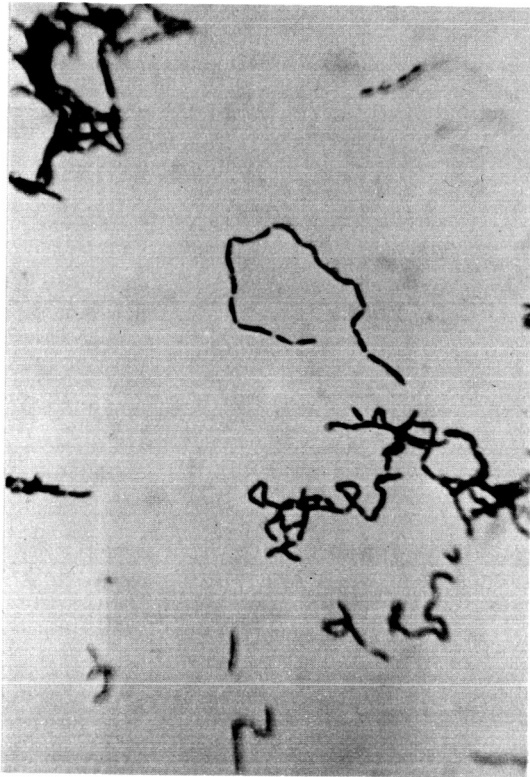


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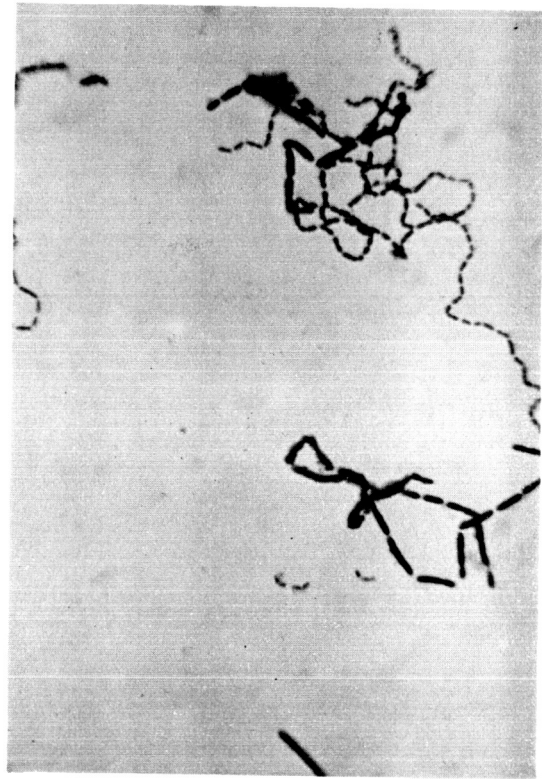


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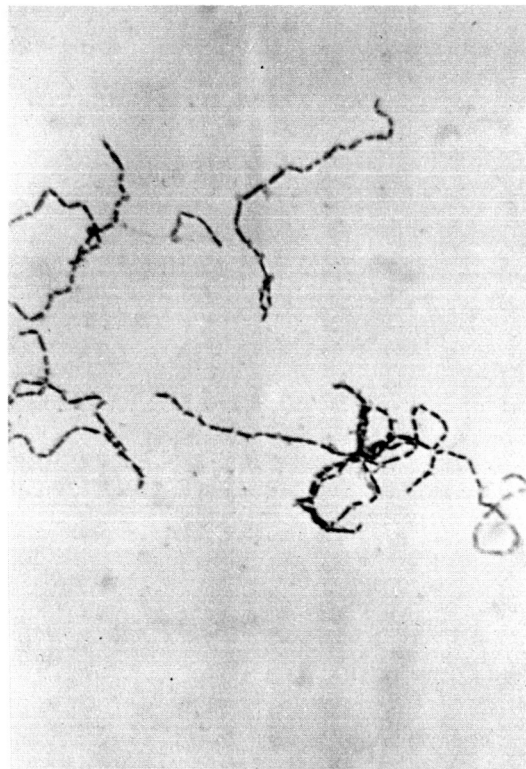
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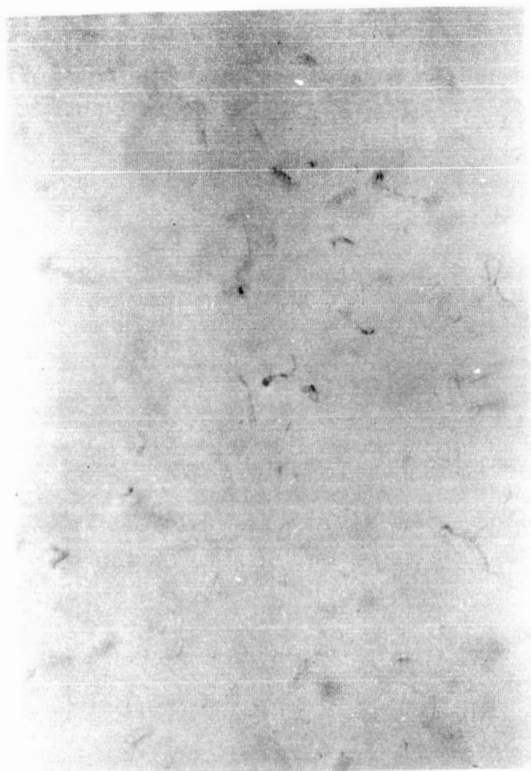


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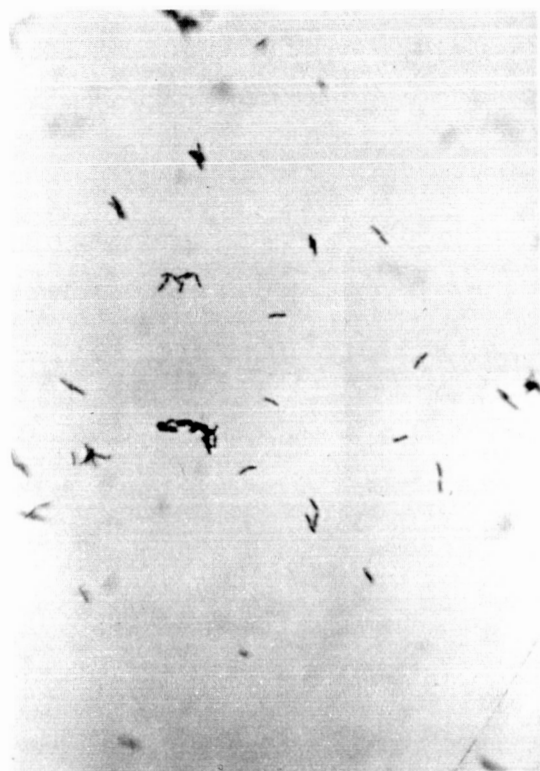


44-Feces-16(9)

Figure 14 --- Concluded



FA-1



FA-2

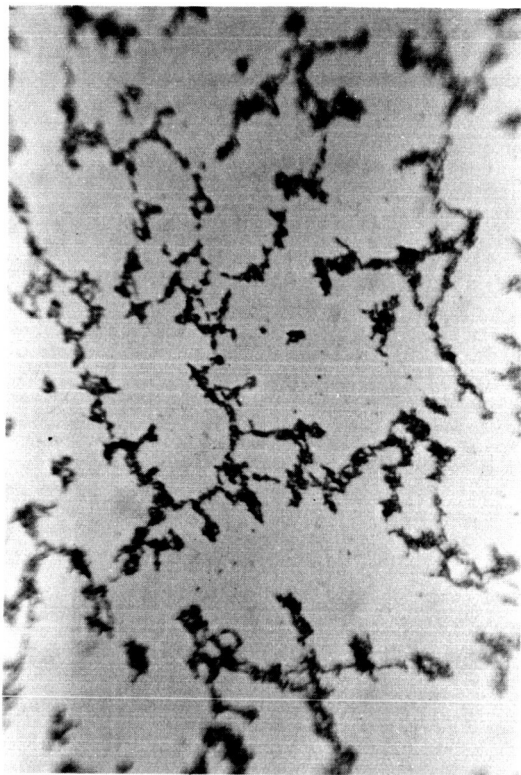


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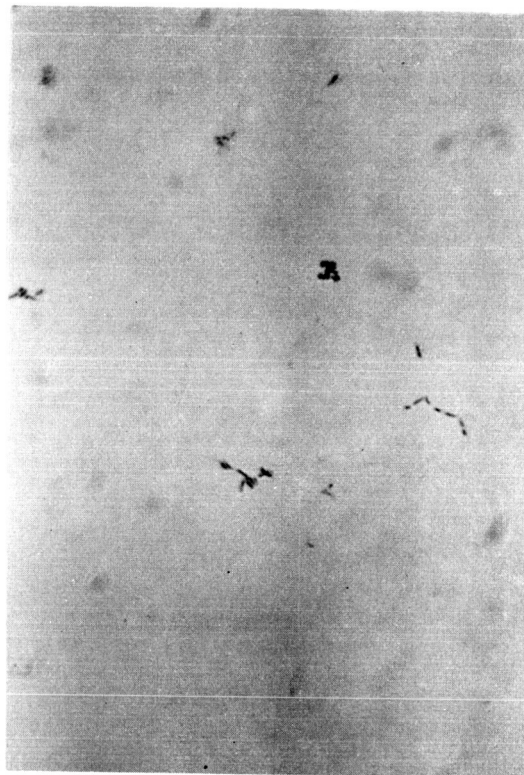


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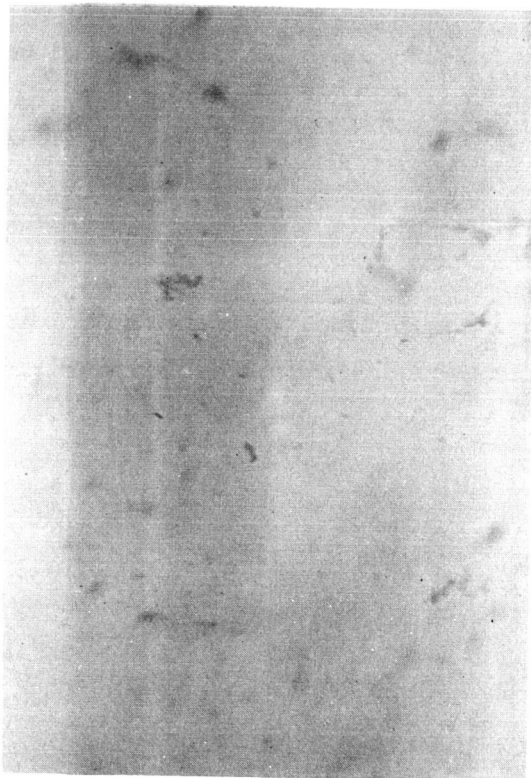
Figure 15. FA Type Cultures



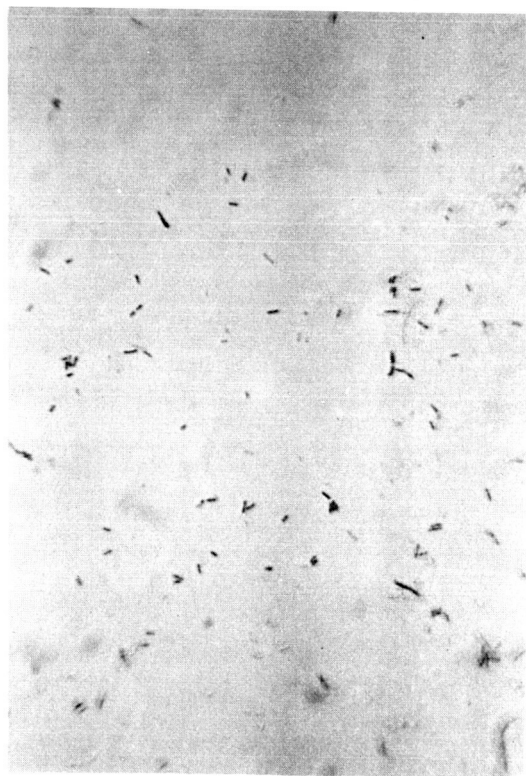
FA-5



FA-6



FA-7



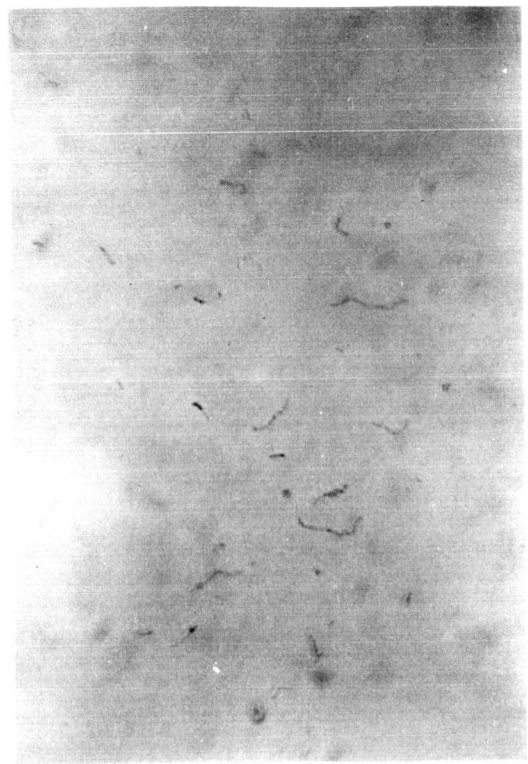
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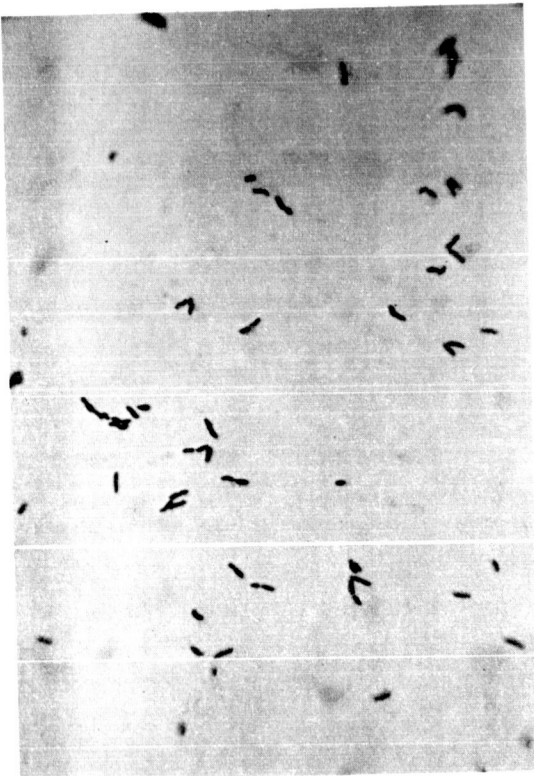




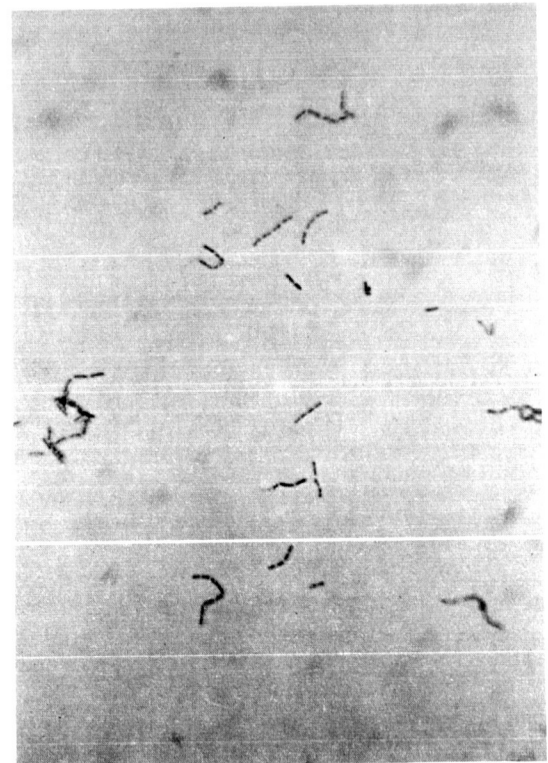
FA-9



FA-10

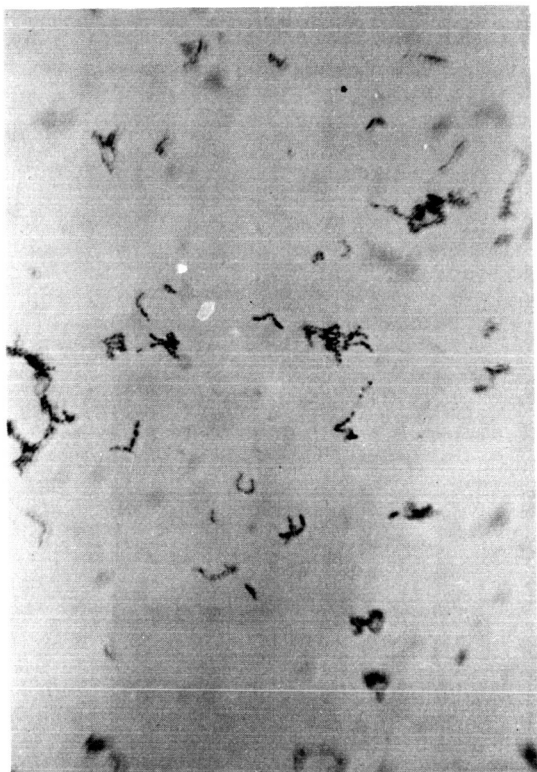


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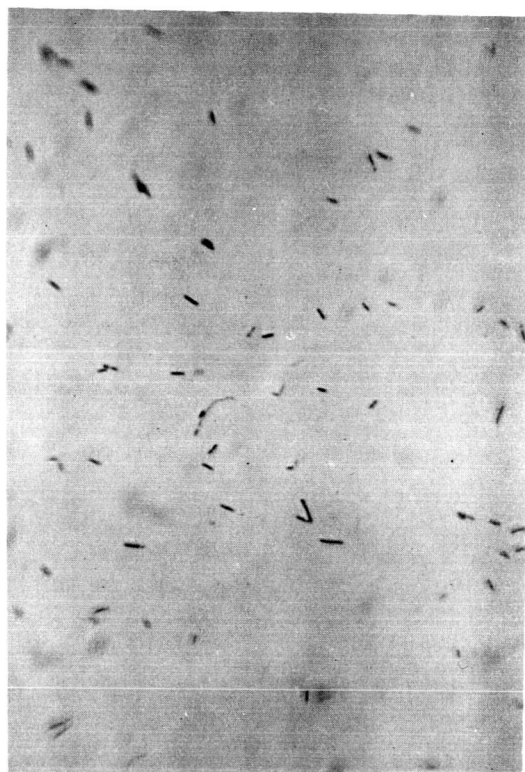


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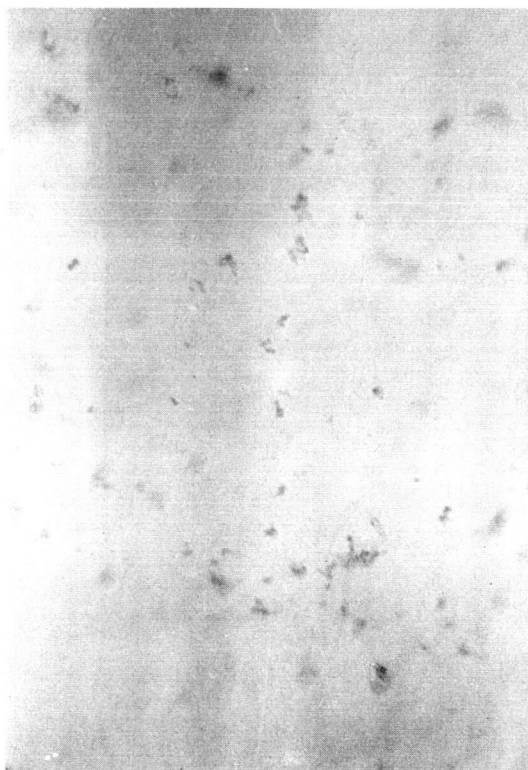
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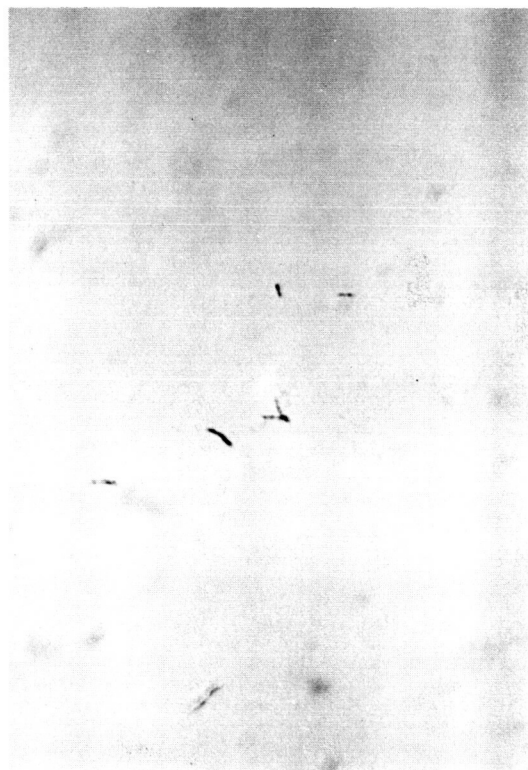
FA-13



FA-14

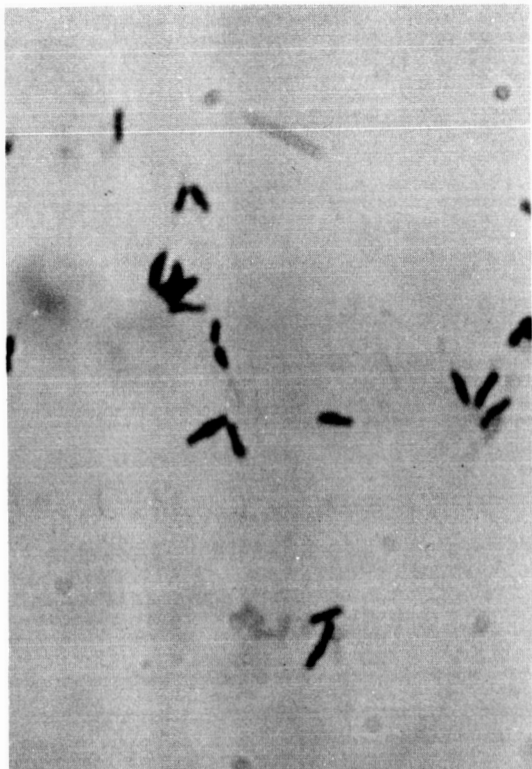


FA-15

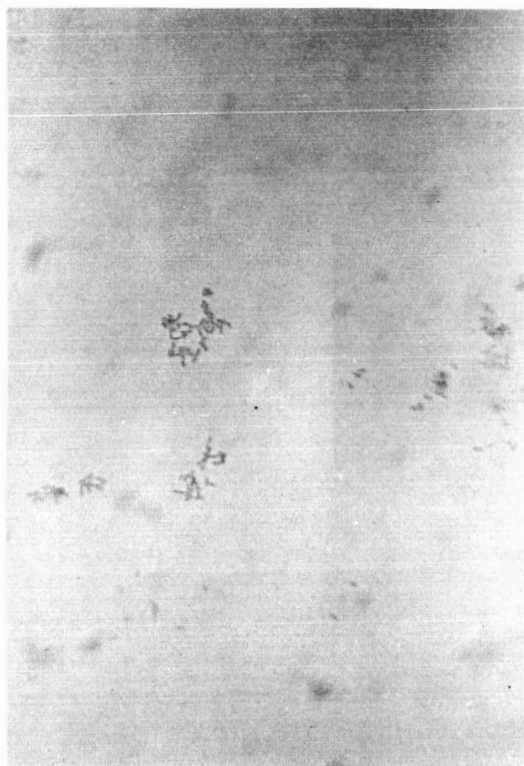


FA-16

Figure 15 --- Continued

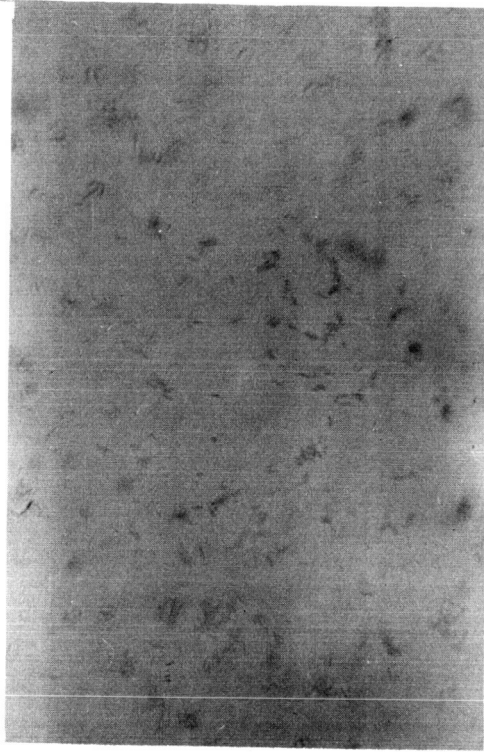


FA-17

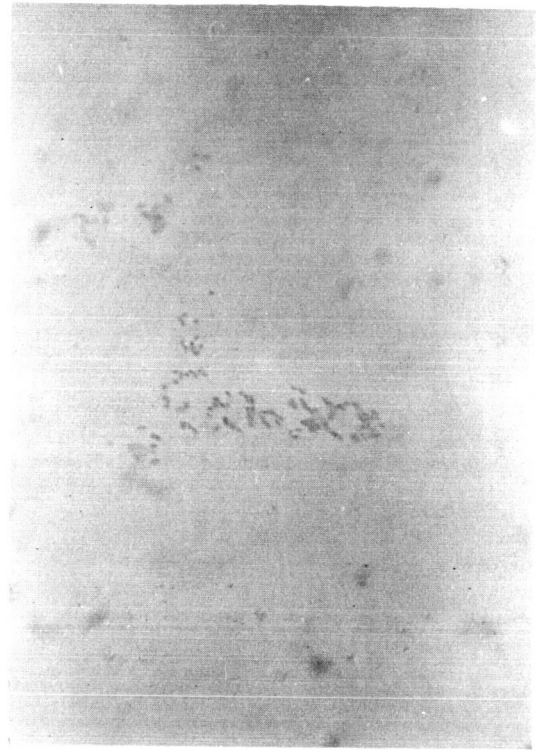


FA-18

Figure 15 --- Concluded



GD-1



GD-2



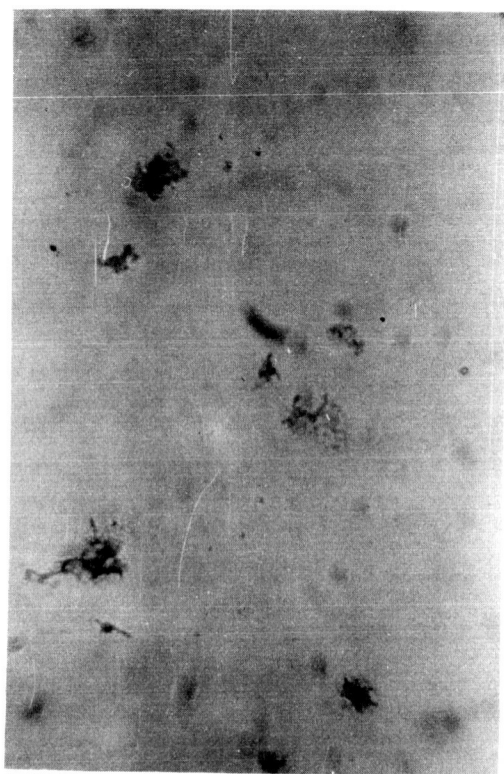
GD-3



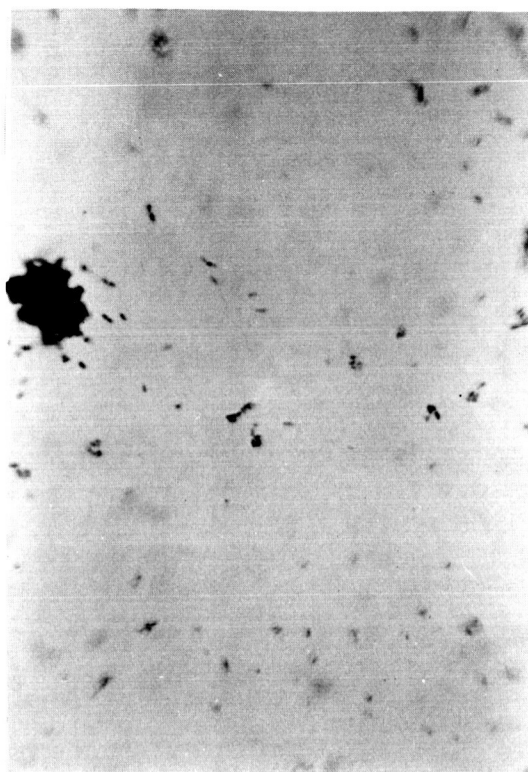
GD-4

Figure 16. GD Type Cultures





GD-5

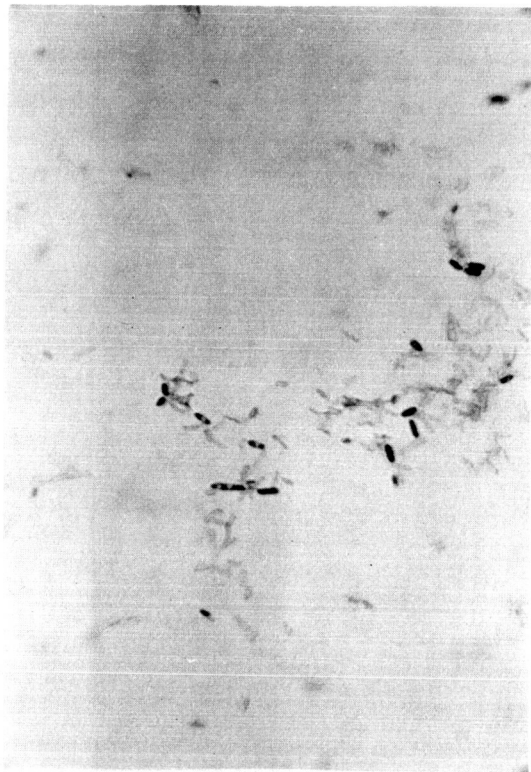


GD-6

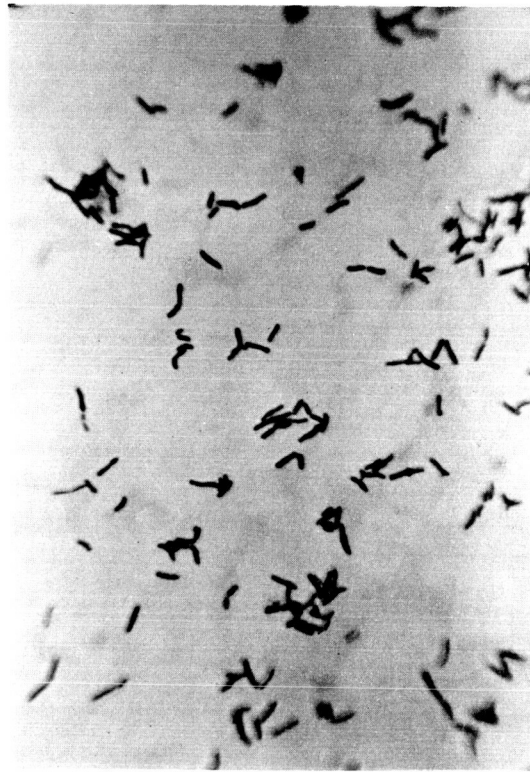


GD-7

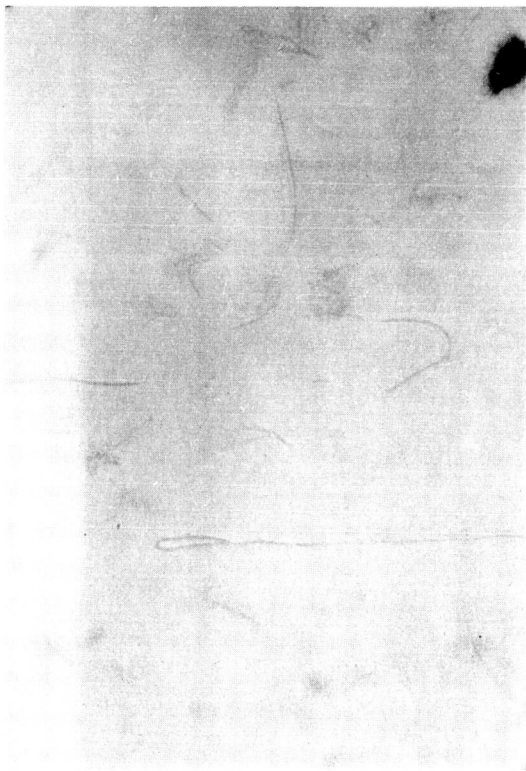
Figure 16 --- Concluded



*Clostridium butyricum*



*Eubacterium limosum*

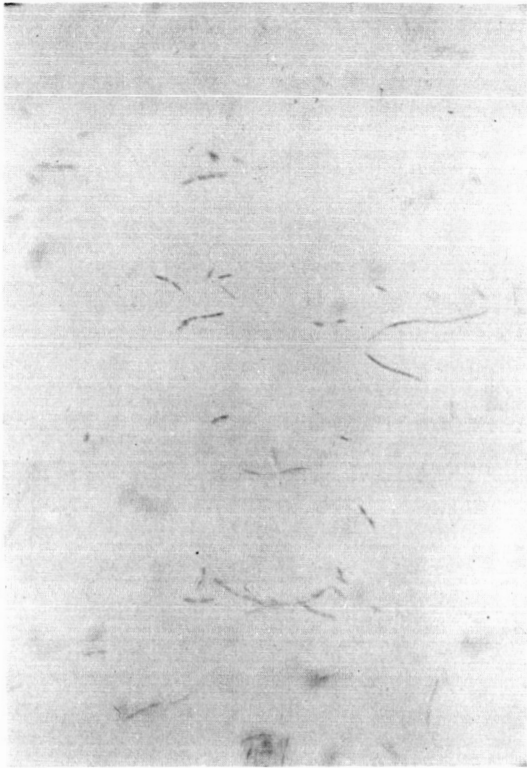


*Fusobacterium polymorphum*

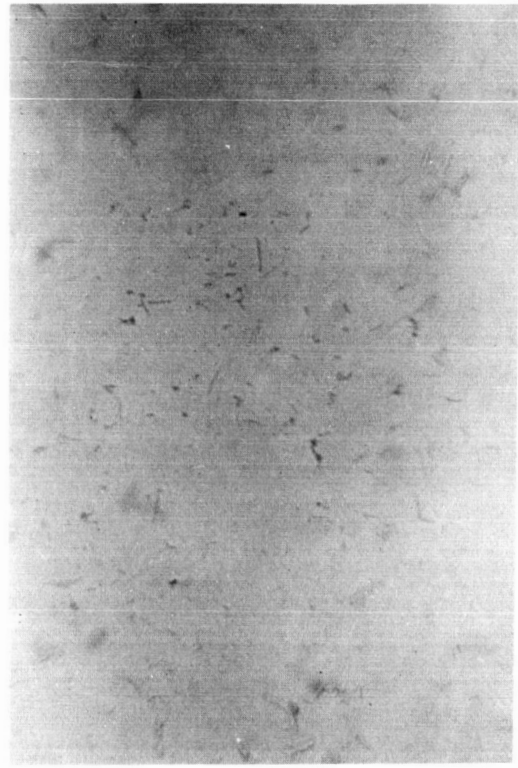


*Lactobacillus bifidus*

Figure 17. Representative American Type Cultures



*Ramibacterium pseudoramosum*



*Sphaerophorus necrophorus*

Figure 17 --- Concluded

## SECTION VI

### CONCLUSIONS

At the beginning of the study, baseline data were obtained from the body areas of all subjects. The bacterial counts obtained at that time were considered to represent normal populations. The effects of confinement and limited personal hygiene on the skin flora and the effect of diet on the intestinal flora were determined from the variations obtained both in the qualitative and quantitative data as the experiment progressed. The data obtained both in the baseline and in subsequent periods differed qualitatively from that reported in the literature<sup>(3)</sup>. In particular, members of the corynebacteria predominated on most body areas at all sampling periods. The literature<sup>(3)</sup> would seem to indicate staphylococci are the predominating microorganisms. The reported predominance of the staphylococci may be due to the ease of culture and viability of this species. The number of organisms found varied among individuals, as well as selectively on different body areas of each individual. Sweating appears to result in a transitory increase in the resident microbial flora.

There was a general rise in microbial levels until approximately the 21st to the 25th day, which was proportional to the time the subjects were confined in the LSSE. This increase in microbial population did not seem to be selective, since the bacteria indigenous to a particular area generally increased proportionately. The exception was in the appearance of members of the Enterobacteriaceae on areas where they do not normally occur. This was particularly evident when the space suit was worn, since gram-negative rods were consistently cultured from the axillary areas of the subjects. Those organisms gradually declined and disappeared after the suits were taken off.

Isolates considered to belong to the genus *Candida* were cultured from 50% of the fecal specimens, from the groin areas of many of the subjects, and from their throats and tongues. The level of occurrence exceeded that found in the literature but agreed with information gained from previous studies in our laboratory.



To determine if the microbial character of the body is bilateral, both the left and right groin areas of each of the four subjects were sampled 26 times during the third experimental period. When the numerical data from these areas were averaged, excellent agreement occurred between the recovery from left and right groin on the four subjects. However, the qualitative differences were marked, since E. coli was isolated only from the right groin of one subject and from the left groin of another subject, while a third subject alternated the recovery of Aerobacter species between the left and right groin. The fungal recovery was more consistent, with the subject carrying Trichosporon on both the left and right groin at the same sampling period.

The anaerobic bacteria recovered from body areas consisted mainly of members of the Peptococcus species. They appeared to be indigenous to the groin and anal area, as well as to the gingival area.

The body maintains a homeostatic balance by absorption, utilization, generation, and excretion. Most of these functions are intimately related to the gastrointestinal tract. The microbial composition of the fecal material reflects the effectiveness of the absorption and utilization of a particular diet. The activity of this microbial flora is of more than academic interest, since its function seems multifold: (1) it influences the host's susceptibility to enteric infection, (2) it produces large quantities of vitamins, (3) it helps maintain a favorable liver function, and (4) it breaks down complex end-products of metabolism and, in so doing, prevents the accumulation of toxic amines. One of the most important conclusions resulting from this study is the determination of the marked shifts in the nonsporulating fecal anaerobes. Many investigators place all nonsporulating anaerobes into the group Bacteroides and make no differentiation of changes within this group. In this particular study, as in previous studies<sup>(8, 12, 13, 14)</sup>, it was in the marked intergroup shifts of nonsporulating anaerobes as a response to dietary influence that the most marked change occurred. The organisms isolated during the latter portions of the experiment were extremely proteolytic and produced large amounts of gas. This type of fecal flora would be undesirable from the viewpoint of space missions, since any increase in flatus produces physical discomfort and introduces toxic compounds into the environmental control system. The dietary period was not

of sufficient length to allow an evaluation of physical symptomatology, but the *in vitro* analysis of predominating members of the fecal flora leads to the conclusion that a lengthened experimental period might reveal adverse physical symptoms.

The effect of diet on the fecal microflora has usually been studied from the viewpoint of the aerobic flora, rather than the anaerobic flora. In this particular study, there were interesting changes in the aerobic, as well as in the anaerobic flora. The most common trend of thought regarding fecal bacterial populations relates them to the presence or absence of diarrhea. The bacterial species responsible for this condition have been studied exhaustively and the relationship between the coli serotypes 055:B4 and 0127:B8 and the disease seems well established. The data from the present study, as well as that from previous studies<sup>(8, 12, 13, 14)</sup> indicate that either the defined diet or the confinement, or the combination of these factors, allows potentially pathogenic serotypes of *E. coli* to become prevalent. This prevalency, while not linked in every instance to diarrhea, allows a potential source of danger to exist in a closed environment.

During an earlier study, typable strains of *E. coli* were recovered from over 50% of the fecal samples. This greatly exceeds the percent of occurrence found by other research workers. During this experiment, all coli colonies occurring on MacConkey's plates (in the range acceptable to standard methods) were identified at eight sampling periods, with interesting results. In the beginning of the experiment, one subject's coli flora consisted exclusively of nontypable organisms, but by the 16th sampling period more than 50% of the coli isolated were of the enteropathogenic type 0125:B15. This may have been due to the diet and ensuing unstabilized condition in the intestinal tract which allowed a minority of organisms in the flora to become predominant. On another subject, roughly 80% of the colonies typed were Poly A 026:B6 (potentially pathogenic).

The microbial levels in the LSSE, as well as those in the CAF, increased proportionately to the increased levels found on the subjects. There were few bacterial species which were not common to both. These consisted of sporadic isolations of bacilli and nocardia, as well as a few saprophytic members of the yeast group.

The most interesting exchange between man and the environment occurred during the third experimental period when a phage typable strain of Staphylococcus aureus was isolated from the CAF and then this member of the phage complex 52/52A/80/81 was subsequently isolated at 19 of the 26 sampling periods. It spread from the floor of the personal hygiene area to the table, to the gingiva of one subject, feces and gingiva area of another subject, as well as to the bed of the first subject. In this particular experiment, no demonstrable illness resulted from the carriage of this potentially pathogenic Staphylococcus aureus type. However, it is interesting to postulate that while these particular subjects were not exposed to stress and were in excellent condition, the same medical outcome could not necessarily be expected under the real stress of space travel.

Transference between the environment and the man has been demonstrated by the Staphylococcus aureus transference described above, and transference between men probably occurred with candida as well as with members of the Enterobacteriaceae species. For example, subject 44 carried aerobacter as a member of his indigenous fecal flora, subsequently, aerobacter was isolated from the feces of subject 41, 42, and 43. Providence was repeatedly isolated from subject 37 and only once from subject 39. Rhodotorula appeared at sampling period 1 on the tongue of subject 40 and was isolated frequently from this subject and subsequently from subjects 37, 38, and 39.

The results of the statistical treatment of the numerical data of this and the previous experiment serve as a useful tool in formulating biomedical criteria for personal hygiene. We suggest that the man should wash every 10 days to maintain his normal microbial levels, with particular attention paid to the axillar, groin, glans penis, and anal areas. The study showed that the groin is an excellent indicator area, signaling deterioration in standards of personal hygiene. Therefore, microbial monitoring of the groin would indicate any necessity to increase the frequency of the washing schedule. The environmental area should be cleaned at the same time intervals as the man. Particular attention should be directed to the personal hygiene area. Fecal material should be handled in a manner which will allow no accidental contamination of the environment. In addition, attention should be directed to those materials used to clean either the man or the cabin. After use, these materials should be bagged or so handled that they are isolated from the

environment. The discarded food wrappers or containers should be considered potentially dangerous since they offer a food source to the bacteria present in the environment. Common equipment handled by more than one individual (i.e., communication equipment, food preparation center, beds, and tables) should be cleaned on a predetermined basis to prevent the buildup of bacteria on their surfaces. The necessity for these sanitation procedures is based on the fact that the stressed astronaut will be more susceptible to infection than the subjects tested. Every potential source of bacterial contamination must be monitored, since bacteria which are indigenous to one individual are not necessarily harmless when implanted or transferred to another individual.

One of the significant contributions of this study to the field of bacteriology and nutrition is the generic identification of the predominating nonsporulating fecal anaerobes. The importance of this identification is related to the drastic changes which occurred not in the numbers, but in the kinds of nonsporulating anaerobes predominating in the higher dilutions of fecal material of men on defined diets.

**APPENDIX**  
**TABULATION OF RESULTS**

TABLE 1. EXPERIMENTAL DESIGN - Experiment X

Area	Date	CONTROLLED ACTIVITY FACILITY														EVALUATOR														CONTROLLED ACTIVITY FACILITY																																		
		M							T							S							M							T							S							M							T							S						
		20	21	22	23	24	25	26	27	28	29	30	10/1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31																					
Microbiology: Skin Feces																																																																
Sweat Test																																																																
Diets: Fresh Matching Menu Cycle																																																																
	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2														
MA-10 Suit (Subjects 38 and 40)																																																																
Body Areas: A B																																																																
Feces: Subject 37 Subject 38 Subject 39 Subject 40																																																																

September 19 Arrival and briefing of subjects  
 September 20 Controlled Activity Facility (B-248)  
 September 28 All subjects on day shift schedule  
 October 4 Evaluator B-824  
 October 7 "Complete isolation"  
 October 9 Subjects on two-shift schedule  
 October 18 Controlled Activity Facility (B-248)  
 October 26 All subjects on day shift schedule  
 November 1 Debriefing and leaving of subjects  
 A Areas are: Nose, throat, gingiva, axilla, groin, glans penis, anal, and toes  
 B Areas are: Eye, ear, scalp, umbilicus, forearm, and tongue

TABLE 1 ---- Continued ---- Experiment Xa

	CAF					Evaluator						
	11/3	11/4	11/5	11/6	11/7	11/8	11/9	11/10	11/11	11/12		
A Areas	1		2			3		4		5		
B Areas			1									
Subject A		1					2		3			
B			1				2			3		
C		1										

Feces

	Evaluator										
	11/13	11/14	11/15	11/16	11/17	11/18	11/19	11/20	11/21	11/22	11/23
A Areas			6		7		8				9
B Areas			2								3
Subject A				4		5			6		
B				4		5			6		
C				2				3			

Feces

A Areas: nose, throat, gingiva, axilla, groin, glans penis, anal, toes, room areas  
 B Areas: eye, ear, scalp, forearm, umbilicus, tongue

TABLE 1 ---- Concluded ---- Experiment XI

Day of Experiment	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30
DATE	2/28	3/1	3/2	3/3	3/4	3/5	3/6	3/7	3/8	3/9	3/10	3/11	3/12	3/13	3/14	3/15	3/16	3/17	3/18	3/19	3/20	3/21	3/22	3/23	3/24	3/25	3/26	3/27	3/28	3/29
Body Sample	1	2	3					4	5	6					7	8	9					10	11	12				13	14	
Fecal Sample																														
41	1	2					3	4							5			6			7	8						9		
42	1	2					3								4			6				7	8					9		
43	1	2					3	4							5	6					7	8						9		
44	1	2					3	4							5	6					7	8						9		
Confinement	CAF																													
Diet	Control										Contingency										Control									
Wipes	NONE										DRY																			

Day of Experiment	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60
DATE	3/30	3/31	4/1	4/2	4/3	4/4	4/5	4/6	4/7	4/8	4/9	4/10	4/11	4/12	4/13	4/14	4/15	4/16	4/17	4/18	4/19	4/20	4/21	4/22	4/23	4/24	4/25	4/26	4/27	4/28
Body Sample	15					16	17	18				19	20	21						22	23	24					25	26		
Fecal Sample																														
41	10					11	12					13	14							15										
42	10											11	12							13										
43						10			11					12						13							14			
44						10		11					12	13						14							15			
Confinement	CAF										LSSE										CAF									
Diet	Control										Gemini										Contingency									
Wipes	DRY										41 Wet (4/day) 42 None										43 Wet (2/day) 44 Wet (2/day)									

• Sweat Test  
\* Shower



TABLE 2. LIST OF PRIMARY CULTURE MEDIA FOR EACH BODY AREA

## Aerobic Samples

	Scalp	Ear	Eye	Nose	Mouth	Gingiva **	Throat	Axilla	Forearm	Umbilicus	Groin	Glans penis	Anal fold	Feces	Toes	Tongue
Actinomycete Agar (c)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
2 Blood Agar Plates (d)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
PPLO Agar (c)*	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Phytone Yeast Extract Agar (d)	X	X		X	X		X	X	X	X	X	X	X	X	X	X
Mitis Salivarius Agar (e)				X	X	X	X							X		X
MacConkey's Agar (e)											X	X	X	X		
PEA***						X					X			X		
Aerobic Dilution Series	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

## Anaerobic Samples

	Scalp	Ear	Eye	Nose	Mouth	Gingiva **	Throat	Axilla	Forearm	Umbilicus	Groin	Glans penis	Anal fold	Feces	Toes	Tongue
Blood Agar Plate (d)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Chocolate Agar (d)				X	X	X	X					X				X
Rogosa's Media (d, e)					X	X	X									
Dilution Series	X	X	X	X	X	X	X	X	X	X	X	X	X	X <sup>(a)</sup>	X	X
Agar Shakes	X	X	X	X	XX	XXX	XX	X	X	X	X	X	X	XXX <sup>(b)</sup>	X	X
Pour Plates					X	X	X					X	X	XX <sup>(b)</sup>		
Counting Plates														X <sup>(b)</sup>		

\* One time per week for body areas

\*\* Dental instruments used for obtaining sample

\*\*\* Phenyl Ethyl Alcohol Agar on Experiment XI Only

(a) Gall's Broth

(b) Gall's Agar

(c) Difco Laboratories

(d) Baltimore Biological Laboratory

(e) Albitri Laboratories, Inc.

TABLE 3. SCREEN TEST FOR PREDOMINATING OBLIGATE AND FACULTATIVE ANAEROBIC FECAL BACTERIA

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
FA-1	slender gram positive rod singly and in chains; distinct rods uniformly spaced	very fine colonies; obligately anaerobic	heavy turbidity with slime developing	4+	4+	4+	2+	+	delayed ARC* with proteolysis	no liquefaction	7.0
FA-2	slender gram positive rod in chains, with tadpole	diffuse colonies; obligately anaerobic	heavy with slime	4+ with silky turbidity 4+ slime	3+ with silky turbidity 3+ slime	3+ with silky turbidity 3+ slime	± +	± +	delayed ARC* with proteolysis	no liquefaction	6.4
FA-3	medium to small gram negative elongate pointed rods in pairs	diffuse growth; heavy gas; obligately anaerobic	heavy with slimy sediment	4+ slimy sediment 4+ black sediment	4+ slimy sediment 4+ black sediment	4+ slimy sediment 4+ black sediment	4+ slimy sediment 4+ black sediment	4+ slimy sediment 4+ black sediment	delayed ARC* with proteolysis and gas	no liquefaction	7.5
FA-4	slender gram positive, sometimes slightly curved rod, singly	small colonies; obligately anaerobic	moderate turbidity	4+ slime 4+ slime	4+ slime 4+ slime	4+ slime 4+ slime	2+ sediment 2+ sediment	2+ sediment 2+ sediment	ARC* strong delayed proteolysis	no liquefaction	5.6
FA-5	short, medium slightly curved gram positive rod, singly; often developing clusters	medium colonies; obligately anaerobic	moderate turbidity	4+ slime 4+ slime	4+ slime 4+ sediment	4+ slime 4+ sediment	4+ slime 4+ slime	± ±	delayed ARC* with proteolysis	no liquefaction	5.5-5.8
FA-6	gram positive medium rods, tending to form clusters some slightly curved	medium colonies; obligately anaerobic	clear slimy sediment	4+ slime 4+ slime	4+ slime 4+ slime	4+ slime 4+ slime	3+ slime 4+ slime	+ slight slime + slight slime	ARC*	no liquefaction	6.6

Results obtained under NASA contract NASw-738, "Study of the Normal Fecal Bacterial Flora of Man."  
\* Acid Reduced Curt

TABLE 3 --- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
FA-7	small gram negative slender rod, tendency towards bipolar staining	fine colonies; obligately anaerobic	moderate turbidity slime	4+ slime  4+ slime	4+ slime  4+ slime	4+ slime  4+ slime	+  + slime	+  +	ARC* delayed proteolysis	no liquefaction	6.6
FA-8	tiny gram negative slender rods, slightly curved	fine colonies; obligately anaerobic	clear with sediment	+  3+	+  3+	+  3+	+  3+	+  3+	partial reduction orange color	no liquefaction	6.9
FA-9	medium to large pleomorphic gram positive rod in pairs and short chains; chain has characteristic hooked or loop shape - older cultures form heavy gram positive aggregation	haze; obligately anaerobic	moderate turbidity	3+ slight slime  3+ moderate slime	3+ slight slime  3+ moderate slime	+ slime  3+ slime	+ slime  + slight slime	clear with slight slime  +	delayed ARC* with + proteolysis	no liquefaction	7.0
FA-10	very small gram positive rods in chains with a tendency for bipolar staining, sometimes slightly pointed	fine colonies; obligately anaerobic	heavy with floccular sediment	4+ fluffy sediment  4+ sediment	4+ fluffy sediment  4+ sediment	4+ fluffy sediment  4+ sediment	3+  4+ sediment	+ sediment  4+ sediment	delayed ARC* with proteolysis	no liquefaction	6.7
FA-11	medium short gram positive rods, some slightly curved, older cultures tend toward gram positive aggregation	fine colonies; obligately anaerobic	heavy turbidity	3+  3+ sediment	3+  3+ sediment	3+ sediment  3+ sediment	3+  3+ sediment	+ sediment  clear with slight sediment	ARC* with proteolysis	no liquefaction	6.5
FA-12	gram positive tiny pointed rods in chains with many coccoid forms	medium colonies; obligately anaerobic with slight gas	heavy with slime	3- slime  3- slime	3- slime  3+ slime	+ with slime  3+ slime	+ slime  + slime	+ slime  + slime	delayed ARC* with proteolysis	no liquefaction	7.2

TABLE 3 --- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
FA-13	small gram negative cocci in masses	fine colonies; heavy gas; obligately anaerobic	moderate turbidity	3+ gas black slime	3+ gas black slime	3+ gas black slime	3+ gas black slime	3+ gas black slime	Reduced	no liquefaction	6.7
FA-14	gram negative rods, long slender with gram positive areas	tiny colonies; obligately anaerobic with heavy gas	heavy turbidity gas	4+ slight slime gas	4+ slight slime	+	±	±	Reduced, whey caramelization	no liquefaction	6.75
FA-15	short fat gram negative rod, singly and in pairs; some with pointed ends	delayed haze; heavy gas; obligately anaerobic	heavy with slight slime	4+ slight slime	4+ slight slime	+	2+ slight slime	±	delayed ARC* with whey	no liquefaction	6.7
FA-16	gram positive pleomorphic rods; some curved and some tadpole forms	haze with anaerobic collar	heavy with slime	+ curly slime 3+ slime	+ curly slime 3+ slime	4+ black slime	4+ slime	±	ARC*	no liquefaction	6.8
FA-17	large gram positive rod singly and in pairs forming palisades and V's	fine colonies; obligately anaerobic; slight gas, variable	slight with finely granular sediment and side growth	clear with finely granular sediment	clear with finely granular sediment	clear with finely granular sediment	clear with finely granular sediment	clear with finely granular sediment	ARC* with	no liquefaction	6.6
FA-18	gram positive long slender rods, irregular staining	fine colonies; obligately anaerobic	slight with slime	± moderate slime	± moderate slime	± moderate slime	± moderate slime	± moderate slime	ARC* delayed	no liquefaction	6.3 to 6.6

TABLE 3 --- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
GD-1	short gram negative rod in pairs and chains, some pointed	fine colonies; heavy gas; obligately anaerobic	heavy floccular sediment	4+ with slime	4+ with slime	4+ with slime	2+ with slime	1+ with slime	delayed ARC* with proteolysis	black bottom no liquefaction	6.7
GD-2	gram negative short rod in pairs	small colonies; obligately anaerobic	moderate with floccular slime	4+ with heavy slime	4+ with heavy slime	4+ with heavy slime	4+ with heavy slime	3+ with floccular slime	ARC* with proteolysis	no liquefaction	6.2 6.4
GD-3	gram negative pointed rods	tiny colonies; obligately anaerobic	moderate with sediment sometimes fluffy	2+ with slime	2+ with slime	2+ with slime	2+ with slime	2+ with slime	reduced	no liquefaction	6.8
GD-4	gram negative slender rods in pairs some pleomorphic	tiny colonies; heavy gas; obligately anaerobic	moderate with granular sediment, sometimes dark	4+ with slime and gas	4+ with slime and gas	4+ with slime and gas	4+ with slime and gas	3+ with slime	delayed ARC* with slight proteolysis	no liquefaction	6.3 6.4

Results obtained under contract AF33(615)-1748, "Determination of Aerobic and Anaerobic Microflora of Human Feces."

TABLE 3 --- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
GD-5 and GD-5a	gram + medium rods in short chains	small colonies, obligately anaerobic	clear to moderate with balls of sediment	4+ with granular sediment or slime	4+ with granular sediment or slime	4+ with granular sediment or slime	4+ with granular sediment or slime	2+ with granular sediment	ARC+ with proteolysis	no liquefaction	6.6 GD-5a 6.2 to 6.4
				4+ with slime or granular sediment sometimes black	4+ with slime or granular sediment sometimes black	4+ with slime or granular sediment sometimes black	4+ with slime or granular sediment sometimes black	3+ with slime or granular sediment sometimes black			
GD-6	gram negative short pleomorphic rods in pairs some pointed	tiny colonies, heavy gas, obligately anaerobic	slight to moderate with slimy sediment	3+ with granular sediment	3+ with granular sediment	3+ with granular sediment	3+ with granular sediment	+ with slimy sediment	delayed ARC* with proteolysis	no liquefaction	5.9
				4+ with brown slime	4+ with brown slime	4+ with brown slime	4+ with brown slime	3+ with brown slime			
GD-7	gram + short pleomorphic rods in pairs some pointed	tiny colonies, heavy gas, obligately anaerobic	4+ with dark slime	4+ with slime and heavy gas	4+ with slime and heavy gas	4+ with slime and heavy gas	3+ with heavy slime and gas	3+ with heavy slime	reduced	no liquefaction black bottom	6.8
				4+ with heavy black slime	4+ with heavy black slime	4+ with heavy black slime	4+ with heavy black slime	4+ with heavy black slime			

TABLE 3 ---- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Destrin	Blank	Litmus Milk	Gelatin	pH
FN-1	gram positive pointed rods in pairs and short chains	fine colonies facultatively anaerobic	heavy with slime	4+ slime 4+ slime	4+ slime 4+ slime	3+ slime 4+ slime	3+ slime 4+ slime	3+ slime 4+ slime	delayed ARC*	no liquefaction	6.7
FN-2	gram positive coccobacillus pairs and chains	medium colonies facultatively anaerobic	clear with growth on sides and white sediment	3+ granular sediment 3+ granular sediment	3+ granular sediment 3+ granular sediment	3+ granular sediment 3+ granular sediment	+ granular sediment 3+ granular sediment	± + with sediment	ARC* with	no liquefaction	6.5
FN-3	small round cocci in short chains becoming less discrete with age	discrete colonies with heavy gas facultatively anaerobic	moderate with white sediment	3+ granular sediment 4+ granular sediment	3+ granular sediment 4+ granular sediment	4+ sediment 4+ granular sediment	3+ 3+ granular sediment	± ±	ARC* with proteolysis	no liquefaction	6.4
FN-4	gram positive elongate cocci in short chains	fine colonies facultatively anaerobic	moderate	4+ slime 4+ slime	4+ slime 4+ slime	3+ slime 4+ slime	3+ slime 4+ slime	3+ slime 4+ slime	delayed soft ARC*	no liquefaction	6.5
FN-5	gram positive diplococci in pairs and short chains; pleomorphic	fine colonies; facultatively anaerobic	moderate with floccular sediment	3+ floccular sediment 4+ floccular sediment	3+ floccular sediment 4+ floccular sediment	3+ floccular sediment 4+ floccular sediment	3+ floccular sediment 4+ floccular sediment	+ sediment + sediment	ARC* with slight proteolysis	no liquefaction	7.3 to 7.7

TABLE 3 --- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
PS <sub>1</sub>	gram positive cocci in short chains	tiny colonies with gas facultatively anaerobic	heavy with slime	3+  4+ slime sometimes black	3+  4+ slime sometimes black	3+  4+ slime sometimes black	+  2+ slime sometimes black	+  + slime sometimes black	delayed ARC	no liquefaction	7.5 to 7.8
PS <sub>2</sub>	gram positive cocci in short chains	tiny colonies with gas facultatively anaerobic	moderate with slime	3+ slime  4+ slime	3+ slime  4+ slime	3+ slime  4+ slime	+ slime  4+ slime	+ slime  + slime	ARC; slight proteolysis	no liquefaction	6.8 to 7.0
PS <sub>3</sub>	gram positive cocci in chains	small colonies facultatively anaerobic	heavy with floccular sediment	3+ sediment  4+ sediment	3+ sediment  4+ sediment	3+ sediment  4+ sediment	2+ sediment  3+ slime	+ sediment  + slime	delayed ARC	no liquefaction	6.4 to 6.6



TABLE 3 --- Continued

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Litmus Milk	Gelatin	pH
CT-1	tiny gram negative cocci in clusters	fine colonies with gas, obligately anaerobic	moderate with black granular sediment and gas	+ with dark granular sediment and gas	+ with dark granular sediment and gas	+ with dark granular sediment and gas	+ with dark granular sediment and gas	+ with dark granular sediment and gas	reduced with black bottom	no liquefaction black bottom and gas	7.5
CT-2	gram positive large pointed rods in chains	small colonies heavy gas, obligately anaerobic	heavy with granular sediment	3+ with slime and side growth	3+ with slime and side growth	3+ with slime and side growth	+ with slime and side growth	+ with silky slime and side growth	ARC* with proteolysis and whey	no liquefaction	7.25
CT-3	gram positive slender rods, some in chains, some slightly curved	very fine colonies; obligately anaerobic	heavy with slight gas	4+ with slime and gas	3+ with slime and gas	+ with slime	4+ with heavy slime	+ with slight slime	ARC* with delayed proteolysis	no liquefaction	5.6

Results obtained under Contract AF29(600)-4124, "Study of Bacterial Flora of Alimentary Tract of Chimpanzees."

TABLE 3 --- Concluded

Type Culture	Morphology	Agar Shake	Broth	Glucose	Sucrose	Lactose	Dextrin	Blank	Likmus Milk	Gelatin	pH
CN-1	gram positive rods, some slightly curved, some ovoid in chains	very fine colonies facultatively anaerobic	slight with slime (dark?)	3+ with flocculant granules and side growth	3+ with flocculant granules and side growth	+ with slight slime	3+ with flocculant granules and side growth	+ with slight slime	ARC*	no liquefaction	5.8
							3+ with flocculant granules and side growth	+ with slight slime			
CN-2	gram positive rods some in pairs; various sizes	small colonies facultatively anaerobic	slight with slime	1+ with granular slime	1+ with granular slime	1+ with granular slime	1+ with granular slime	1+ with granular slime	reduction	no liquefaction	7.3
							1+ with granular slime	1+ with granular slime			

TABLE 4. ROOM AREA COUNTS - Experiment X

Sampling Period	Preentry Evaluator											
	1	2	3	4	5	6	7	8	9	10	11	12
Eating/Fore Table Aft Table					6 1	51 330	55 658	111 72	284	140		
Table	35	45	17	80							85	117
Floor - Personnel Hygiene Area	43	60	85	186	16	228	449	56	423	336	250	192
Bed	21	47	9	42	2	235	485	391	232	152	58	250
	CONTROLLED ACTIVITY FACILITY*					EVALUATOR			CONTROLLED ACTIVITY FACILITY			

\*CAF cleaned prior to subjects entering.

TABLE 4 --- Continued --- Experiment Xa

Sampling Period	CAF		Evaluator						
	1	2	3	4	5	6	7	8	9
Tables Fore Aft	6	47	39 300	120 297	23 250	49 178	75 N.S.	39 388	N.S.*
Floor-Personal Hygiene Area	7	81	200	163	Spr.**	147	192	94	
Bed	8	47	270	352	160	99	N.S.	140	

\*NS = No sample

\*\*Spr. = Spreader

TABLE 4 ---- Concluded ---- Experiment XI

	CAF Uncleaned			Post Clean CAF	CAF											
	1	2	3		4	5	6	7	8	9	10	11	12	13	14	15
Sampling Period																
Table	49	49	172	3	115	69	64	177	197	246	91	215	49	80	21	76
Bed	21	57	78	0	41	99	141	487	240	348	77	179	58	141	49	104
Floor - Personal Hygiene Area	81	89	140	7	168	151	111	205	339	326	103	115	63	149	89	106

	CAF						LSSE Entry	LSSE			Pre Clean CAF	Post Clean CAF	CAF	
	16	17	18	19	20	21		22	23	24			25	26
Sampling Period														
Table	31	39	23	24	18	15	7	42	110	56	0	0	58	65
Bed	65	47	61	24	15	25	NS	56	91	82	1	1	36	169
Floor-Personal Hygiene Area	77	92	61	67	55	196	NS	32	48	80	6	1	28	109

CAF Controlled Activity Facility

LSSE Life Support Systems Evaluator

NS No Sample

TABLE 5. MICROORGANISMS FOUND ON ENVIRONMENTAL SAMPLING  
Experiment X

	1	2	3	4	Preentry Evaluator	5	6	7	8	9	10	11
(1) Bed	staph g neg rod	staph	staph	staph g neg rod	staph	staph	staph	staph Coryne Aerobacter	staph Coryne strep	staph	staph Coryne strep	staph Coryne
(1) Fore Table					staph	staph Coryne	staph strep	staph Coryne				
(1) Aft Table	staph	staph	staph	staph Aerobacter Pseudomonas Achromo- bacter gp.	staph	staph	staph Coryne g neg rod	staph Coryne	staph Coryne	staph	staph Coryne	staph
(1) Floor Personal Hygiene Area	staph	staph Bacillus Citrobacter	staph strep	Coryne Bacillus Staph	staph	staph	staph Coryne g neg rod	staph Coryne Aerobacter	staph strep Coryne	staph Coryne	Bacillus staph Coryne Aerobacter	staph strep Coryne
(2) Telephone Mouthpiece		staph		staph		staph	staph	staph	staph (gray mucoid colony)	staph Coryne	staph	staph Coryne
(2) Personal Hygiene Seat		staph		staph		staph Coryne	staph Bacillus	staph Coryne	staph Coryne	staph Coryne		
(2) Refrigerator Handle		staph		staph		staph Coryne	staph Coryne	staph Coryne	staph	staph		staph Coryne

(1) based on sedimentation plates  
(2) based on swabs of surface

TABLE 5 ---- Continued ---- Experiment Xa

Area	Sampling Period							
	1	2	3	4	5	6	7	8
Fore Table			staph	Bacillus staph Aerobacter	staph	Bacillus staph	staph Coryne. sp.	staph
Aft Table	staph	staph Coryne. sp.	Bacillus staph E. coli Poly A Alcaligenes Pseudomonas	Bacillus staph strep Alcaligenes Pseudomonas	staph Alcaligenes Proteus	Bacillus staph C. striatum Pseudomonas	N. S. *	Bacillus staph
Floor Personal Hygiene	staph	staph Coryne. sp. Gm neg rod	Bacillus staph E. coli Poly A Pseudomonas	Bacillus staph Aerobacter	Proteus Alcaligenes	Bacillus staph Proteus Aerobacter mixed	Bacillus staph strep Coryne. sp.	Bacillus staph strep
Bed	staph C. striatum	staph C. striatum	Bacillus staph E. coli Poly B 086:B7 0124:B17 0128:B12 Aerobacter Alcaligenes	Bacillus staph Aerobacter	Bacillus staph Gm neg rod	Bacillus staph Gm neg rod	N. S.	Bacillus staph C. striatum
Personal Hygiene Seat						C. striatum		

\*NS = No sample

TABLE 5 --- Concluded --- Experiment XI

Sampling Area	Pre-clean CAF			Sampling Period											Post-Clean	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14		
Bed (1)	staph S. aureus Coryne Bacillus	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne strep	staph S. aureus A. niger Pen. sp.	staph S. aureus Coryne Aerobacter	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne		
Table (1)	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	S. aureus	staph S. aureus + Coryne	staph S. aureus A. niger Pen. sp.	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne Flavobacterium	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne		
(1) Floor Personal Hygiene Area	staph S. aureus Coryne Bacillus	staph S. aureus + Coryne Mycococcus	staph S. aureus + Coryne	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus Coryne A. niger Pen. sp.	staph S. aureus Coryne	staph S. aureus Coryne Bacillus	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne Mycococcus		
Microphone Mouthpiece (2)				staph Bacillus Coryne	staph strep Coryne	staph Coryne	staph Coryne	staph	staph S. aureus Coryne	staph Coryne Bacillus	staph	staph	staph	Coryne		
Personal Hygiene Seat (2)	Coryne	staph Coryne	staph Coryne	staph Coryne Bacillus	staph Coryne	Coryne	staph Coryne	staph Coryne	staph Coryne	staph Coryne	staph Coryne	staph Coryne	staph Coryne	staph S. aureus Coryne		

Sampling Area	Sampling Period														Pre-Clean CAF		Post-Clean CAF	
	15	16	17	18	19	20	21	22	23	24	25	26	27	28				
Bed (1)	staph S. aureus Coryne Mycococcus	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne				
Table (1)	staph strep Coryne	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne				
(1) Floor Personal Hygiene Area	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne Bacillus	staph S. aureus + Coryne Bacillus	staph S. aureus + Coryne Bacillus	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne				
Microphone Mouthpiece (2)	Coryne	staph Coryne	staph Coryne	S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne				
Personal Hygiene Seat (2)	staph Coryne	staph Coryne	staph Coryne	staph Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne	staph S. aureus + Coryne				

(1) Based on sedimentation plates  
(2) Based on swabs  
+ = Coagulase positive  
Pen. = Penicillium

\* Cowan, S. T. and Steel, K. J., Manual for the Identification of Medical Bacteria, (Cambridge at the University Press, 1969) p. 61-82.  
Pickett, M. J., and Mancilark, C. R., "Tribe Miniae: An Illaginate Epithet," American Journal of Clinical Pathology, Vol. 43, No. 2, (1965) p. 161-165.



TABLE 6. TOTAL BACTERIAL COUNTS BY BODY AREA FOR EACH CULTURING PERIOD

Experiment X

Subject 37

Body Area	Dilution*	Sampling Period										
		1	2	3	4	5	6	7	8	9	10	11
<u>A Areas</u>												
Nose	10 <sup>-3</sup>	389	~1034	9640	139	11610	204	586	269	1100	344	480
Throat	10 <sup>-4</sup>	300	176	27	37	131	51	23	249	40	27	42
Gingiva	10 <sup>-3</sup>	203	138	34	5290	411	170	45	2070	>5790	31	330
Axilla	10 <sup>-3</sup>	3	4	7	103	780	2060	660	690	710	230	63
Groin	10 <sup>-4</sup>	380	>15440	23	20	>1018	676	1212	365	>1001	301	621
Glans penis	10 <sup>-4</sup>	475	442	2	50	40	>851	787 Spr	27	6	102	92
Anal	10 <sup>-5</sup>	157	tntc	501	~ 658	75	488	431	25	151	192	282
Toe	10 <sup>-5</sup>	419	254	236	7	Evaluator			36	16	83	812

B Areas

Eye	10 <sup>-3</sup>	0						NG				11
Ear	10 <sup>-3</sup>	1						503				0
Scalp	10 <sup>-4</sup>	54						28				12
Forearm	10 <sup>-3</sup>	1						23				1
Umbilicus	10 <sup>-3</sup>	1						1				49
Tongue	10 <sup>-5</sup>	> 60						34				97

Spr. = Spreader

NG = No growth

\*0.1 cc of these dilutions were plated

TABLE 6 --- Continued --- Experiment X

Subject 38

Body Area	Dilution*	Sampling Period										
		1	2	3	4	5	6	7	8	9	10	11
<u>A Areas</u>												
Nose	10 <sup>-3</sup>	120	44	18	4	49	21	34	138	174	19	30
Throat	10 <sup>-4</sup>	173	93	50	1032	32	441	3320	96	3540	155	86
Gingiva	10 <sup>-3</sup>	2900	10	761	>12500	520	3400	210	4100	44	1560	10
Axilla	10 <sup>-3</sup>	1	4	6	0	3	11	6	53	27	N.G.	28
Groin	10 <sup>-4</sup>	168	220	>1016	0	503	~1200	670	~797	240	1040	1770
Glans penis	10 <sup>-4</sup>	10	15	10	N.S.	238	84	171	116	69	>649	177
Anal	10 <sup>-5</sup>	4	32	16	2	1	1	15	9	25	15	tnbc
Toe	10 <sup>-5</sup>	43	77	8	0	Evaluator		456		40	130	302

B Areas

Eye	10 <sup>-3</sup>	0						10				0
Ear	10 <sup>-3</sup>	750						>6000				600
Scalp	10 <sup>-4</sup>	370						260				19
Forearm	10 <sup>-3</sup>	1						3				1
Umbilicus	10 <sup>-3</sup>	1						250				0
Tongue	10 <sup>-5</sup>	>50						31				100

NS = No sample; NG = No growth; tntc = Too numerous to count  
 \*0.1 cc of these dilutions were plated

TABLE 6 ---- Continued ---- Experiment X  
Subject 39

Body Area	Dilution*	Sampling Period										
		1	2	3	4	5	6	7	8	9	10	11
<u>A Areas</u>												
Nose	10 <sup>-3</sup>	64	35	17	141	40	1430	250	102	229	517	2430
Throat	10 <sup>-4</sup>	130	7	37	276	167	160	56	158	70	8	24
Gingiva	10 <sup>-3</sup>	24	12	19	68	45	32	9	79	38	32	8
Axilla	10 <sup>-3</sup>	71	tntc	168	8	242	3140	9500	4200	2450	1300	1130
Groin	10 <sup>-4</sup>	110	42	146	5	352	~490	>1168	2900	550	2380	2930
Glans penis	10 <sup>-4</sup>	32	411	52	0	23	157	112	6	178	192	>113
Anal	10 <sup>-5</sup>	2	32	5	1	30	163	82	162	345	31	313
Toe	10 <sup>-5</sup>	114	tntc	23	35	Evaluator		14	13	138	152	

B Areas

Eye	10 <sup>-3</sup>	0						1				1
Ear	10 <sup>-3</sup>	2560						>817				670
Scalp	10 <sup>-4</sup>	3						6				14
Forearm	10 <sup>-3</sup>	0						10				2
Umbilicus	10 <sup>-3</sup>	1						690				49
Tongue	10 <sup>-5</sup>	tntc						38				34

tntc = Too numerous to count  
\*0.1 cc of these dilutions were plated

TABLE 6 --- Continued --- Experiment X

Subject 40

Body Area	Dilution*	Sampling Period										
		1	2	3	4	5	6	7	8	9	10	11
<u>A Areas</u>												
Nose	10 <sup>-3</sup>	123	66	156	28	16	21	2	41	278	5	13
Throat	10 <sup>-4</sup>	420	7	~638	> 603	191	580	360	310	111	580	9
Gingiva	10 <sup>-3</sup>	0	100	51	7240	~340	970	30	4500	990	283	2
Axilla	10 <sup>-3</sup>	1	135	2080	7	107	224	158	20	9	30	154
Groin	10 <sup>-4</sup>	143	139	~240	3	200	747	150	264	2840	134	524
Glans penis	10 <sup>-4</sup>	0	N. G.	1	0	5	7	6	0	2	5	33
Anal	10 <sup>-5</sup>	0	2	21	746	27	16	6	99	18	6	2
Toe	10 <sup>-5</sup>	8	78	56	102	Evaluator			17	33	6	40

B Areas

Eye	10 <sup>-3</sup>	3						1				0
Ear	10 <sup>-3</sup>	2						6520				4000
Scalp	10 <sup>-4</sup>	3						19				19
Forearm	10 <sup>-3</sup>	1						0				11
Umbilicus	10 <sup>-3</sup>	0						6				1
Tongue	10 <sup>-5</sup>	47						303				96

N.G. = No growth

\*0.1 cc of these dilutions were plated

TABLE 6 --- Continued --- Experiment Xa

Subject A

Body Area	Sampling Period								
	1	2	3	4	5	6	7	8	9
A Areas									
Nose	0	55	300	4	1	17	11	3910	6050
Throat	2000	12500	2160	515	1700	7	165	238	407
Gingiva	6	286	1840	18	3070	363	TNTC	100	N.S.
Axilla	6	550	7220	3270	1130	470	260	1840	720
Groin	2	2410	6220	790	1770	6140	2960	3660	9300
Glans penis	1	30	Spr.	Spr.	Spr.	Spr.	Spr.	Spr.	Spr.
Anal	200	4000	2870	490	480	190	400	150	120
Toe	130	> 5500		Suited			11800	6800	24100
B Areas									
Eye	0					0			1
Ear	90					0			0
Scalp	0					32			2
Tongue	5250					1200			12400
Forearm	10					11			1
Umbilicus	0					3			1

Dilution =  $10^{-4}$ 

N.S. = No sample

TNTC = Too numerous to count

Spr. = Spreader

TABLE 6 ---- Continued ---- Experiment Xa

## Subject B

Body Area	Sampling Period								
	1	2	3	4	5	6	7	8	9
A Areas									
Nose	1780	5	620	5710	3560	3620	3760	140	22
Throat	6960	2170	11200	1800	25400	14100	2110	8000	1600
Gingiva*									
Axilla	7	1160	6720**	4920**	20900	1080	1600	700	5800
Groin	221	370	1490	3720	5750	1400	4960	9750	6950
Glans penis	23	27	92	89	5380	138	17	135	108
Anal	1840	>9000	1890	4320	1890	3630	4640	4040	3500
Toe	1130	2810	Suited				1730	830	4310
B Areas									
Eye	1					0			0
Ear	1050					52			82
Scalp	0					8			N.S.
Tongue	5700					2780			18800
Forearm	0					5			0
Umbilicus	4					0			2920

\* Gingiva - no samples

\*\* Predominately Enterobacteriaceae

\*\*\* Many Enterobacteriaceae

Dilution =  $10^{-4}$

TABLE 6 ---- Continued ---- Experiment Xa

Subject C

Body Area	Sampling Period								
	1	2	3	4	5	6	7	8	9
A Areas									
Nose	3290	230	178	740	61	208	444	780	1640
Throat	2910	3150	23400	860	1800	2690	4620	1650	1110
Gingiva	>12000	TNTC	1410	52300	2740	17000	>6000	2710	N.S.
Axilla	53	2820	2470	2390	810	230	710	99	480
Groin	>7000	3600	780	1530	>5000	1100	2460	7000	12400
Glans penis	28	1660	684	91	33	0	3	0	28
Anal	4570	TNTC	3800	4160	2350	7400	>7000	450	28800
Toe	4050	TNTC	Suited				92	2000	TNTC
B Areas									
Eye	2					0			0
Ear	~4000					9			310
Scalp	3					850			3
Tongue	~6000					4410			25100
Forearm	1					2			0
Umbilicus	0					1			0

Dilution =  $10^{-4}$

TABLE 6 ---- Continued ---- Experiment XI

Subject 41

Body Area	Dilution	Sampling Period												
		1	2	3	4	5	6	7	8	9	10	11	12	13
Groin (left)	10 <sup>-4</sup>	1080	9790	3330	750	2910	1120	1180	2950	2900	850	750	4280	3520
Groin (right)	10 <sup>-4</sup>	3280	-	-	5490	9000	5520	5060	16270	33500	1700	1380	4870	1640
Gingiva	10 <sup>-4</sup>	7000	tntc	193000	-	100	30	30	1359	2190	130	81	1111	285

Body Area	Dilution	Sampling Period													
		14	15	16	17	18	19	20	21	22	23	24	25	26	
Groin (left)	$10^{-4}$	1990	1247	1140	986	1198	2100	520	2950	1950	2270	1970	1070	1840	
Groin (right)	$10^{-4}$	350	583	800	1699	1133	2820	1090	1610	670	650	1790	1090	1790	
Gingiva	$10^{-4}$	24	99	145	1051	828	202	363	566	82	30	750	69	575	

- Confluent growth



TABLE 6 ---- Continued ---- Experiment XI

Subject 42

Body Area	Dilution	Sampling Period												
		1	2	3	4	5	6	7	8	9	10	11	12	13
Groin (left)	10 <sup>-4</sup>	690	2090	1310	310	885	2105	200	8000	4930	40	197	7208	1670
Groin (right)	10 <sup>-4</sup>	1720	-	630	140	567	896	880	10	1600	1316	1290	10260	4160
Gingiva	10 <sup>-4</sup>	450	tntc	81400	11500	780	757	2700	1460	27200	-	-	540	1790

Body Area	Dilution	Sampling Period													
		14	15	16	17	18	19	20	21	22	23	24	25	26	
Groin (left)	10 <sup>-4</sup>	887	1003	3130	1179	1122	578	290	309	704	488	1407	262	443	
Groin (right)	10 <sup>-4</sup>	868	1201	1070	944	1155	929	81	247	1106	267	1153	490	336	
Gingiva	10 <sup>-4</sup>	840	8040	450	1102	1203	6320	405	22	29	10	444	1100	575	

- Confluent growth

TABLE 6 ---- Continued ---- Experiment XI

Subject 43

Body Area	Dilution	Sampling Period												
		1	2	3	4	5	6	7	8	9	10	11	12	13
Groin (left)	10 <sup>-4</sup>	320	1570	7670	1500	3290	5700	1900	6420	4900	3340	1780	6070	2680
Groin (right)	10 <sup>-4</sup>	700	1790	14600	3200	7710	4000	2000	1940	2640	2480	2850	6040	1920
Gingiva	10 <sup>-4</sup>	1970	970	-	764	16	29	3	1185	2661	30	7	114	1096

Body Area	Dilution	Sampling Period													
		14	15	16	17	18	19	20	21	22	23	24	25	26	
Groin (left)	$10^{-4}$	3830	2170	4610	3540	2490	6060	670	2260	1110	3390	6080	520	4810	
Groin (right)	$10^{-4}$	1330	2030	680	690	890	2370	480	2150	6900	2230	1720	2990	3480	
Gingiva	$10^{-4}$	360	824	814	827	1360	890	163	81	374	503	655	307	608	

Confluent growth

TABLE 6 ---- Concluded ---- Experiment XI

Subject 44

Body Area	Dilution	Sampling Period												
		1	2	3	4	5	6	7	8	9	10	11	12	13
Groin (left)	$10^{-4}$	2250	tntc	tntc	8700	13820	10080	50000	55600	13000	22300	9600	13500	66800
Groin (right)	$10^{-4}$	1280	95	tntc	1000	2180	12180	56600	49000	52700	51600	8700	46700	11000
Gingiva	$10^{-4}$	1280	tntc	8400	-	50	50	32	1063	13130	41170	400	130	70

Body Area	Dilution	Sampling Period												
		14	15	16	17	18	19	20	21	22	23	24	25	26
Groin (left)	$10^{-4}$	46600	32300	27200	8740	7820	3560	450	3030	6470	9090	17400	500	5730
Groin (right)	$10^{-4}$	9200	24100	7000	3110	7220	5990	240	1120	10560	6860	22000	1000	4290
Gingiva	$10^{-4}$	900	1030	7290	3210	6250	90	108	52	671	119	276	36	1001

- Confluent growth

TABLE 7. RECOVERY OF ENTEROBACTERIACEAE

Experiment X

Subject 37

Body Area	Sampling Period					
	1	2	3	4	5	6
Groin	E. coli NT		Providencia	Alcaligenes		
Glans penis	E. coli NT	Citrobacter			Providencia	Achromobacter gp.
Anal fold	E. coli	E. coli	E. coli	E. coli NT	E. coli NT	E. coli NT
	Poly A 0127:B8	Poly B NFT				
	0111:B4	E. coli				
	Poly B 0124:B17	Poly B 0124:B17				
	E. coli NT					
Toe	Providencia		Providencia	Providencia		
Feces	E. coli NT	E. coli NT	E. coli NT			E. coli NT

Body Area	Sampling Period				
	7	8	9	10	11
Groin	Providencia Aerobacter	Providencia			
Glans penis	Aerobacter				
Anal fold			E. coli NT	E. coli NT	
Toe					
Feces	E. coli NT Citrobacter	E. coli NT	E. coli NT	E. coli NT	E. coli Aerobacter
Miscellaneous		Gingiva - Achromobacter			Throat - Achromobacter

TABLE 7 ---- Continued  
Subject 38

Body Area	Sampling Period					
	1	2	3	4	5	6
Groin		E. coli NT				
Glans penis					Providencia	
Anal fold						
Toe						
Feces	E. coli NT	E. coli NT	E. coli NT	E. coli NT	E. coli NT	E. coli NT
			E. coli Poly B NFT			
			E. coli Poly B 0119:B14			

Body Area	Sampling Period					
	7	8	9	10	11	
Groin						
Glans penis				Pseudomonas		
Anal fold						
Toe						
Feces		E. coli NT	Aerobacter	E. coli NT	Aerobacter	Aerobacter E. coli

TABLE 7 --- Continued  
Subject 39

Body Area	Sampling Period					
	1	2	3	4	5	6
Groin				Alcaligenes	E. coli NT	E. coli NT
Glans penis	Aerobacter	Providencia Citrobacter E. coli NT Aerobacter	Aerobacter		Aerobacter	Aerobacter Alcaligenes
Anal Fold					E. coli NT	Aerobacter Citrobacter
Toe				Pseudomonas		
Feces	E. coli NT		Aerobacter	E. coli, Poly B NFT Aerobacter	E. coli NT Aerobacter	E. coli NT
Misc.				Axilla - Achromobacter group		

Body Area	Sampling Period				
	7	8	9	10	11
Groin					
Glans penis					
Anal fold			E. coli NT		
Toe					
Feces		E. coli NT	E. coli NT	E. coli NT Aerobacter	E. coli Aerobacter
Miscellaneous	Nose - Flavobacterium				

TABLE 7 --- Continued  
Subject 40

Body Area	Sampling Period					
	1	2	3	4	5	6
Groin		Pseudomonas		Aerobacter		
Glans penis						
Anal fold						
Toe						
Feces	E. coli, Poly A 0127:B8; 0111: B4	E. coli NT Aerobacter	E. coli Poly B NFT	E. coli NT		E. coli NT
Misc.					Nose - Achromobacter	Throat - Pseudomonas

Body Area	Sampling Period			
	7	8	9	10
Groin				
Glans penis	Aerobacter			
Anal fold				
Toe				
Feces	E. coli NT			E. coli NT Aerobacter

E. coli NT

TABLE 7 ---- Continued ---- Experiment Xa

## Subject A

Body Area	Sampling Period				
	1	2	3	4	5
Groin					Alcaligenes
Glans penis			Proteus	Proteus	Proteus E. coli NT
Anal			E. coli NT E. coli Poly B NFT E. coli Poly A 055:B5 0111:B4 026:B6		
Feces	E. coli NT	Aerobacter E. coli Poly A NFT	E. coli Poly A NFT	Aerobacter	E. coli Poly A NFT Aerobacter

Body Area	Sampling Period			
	6	7	8	9
Groin				
Glans penis	Aerobacter	Pseudomonas	Proteus	Pseudomonas
Anal				
Feces	E. coli NT Alcaligenes			

NT = No type

NFT = No further type



TABLE 7 --- Continued

Subject B

Body Area	Sampling Period				
	1	2	3	4	5
Axilla			Aerobacter E. coli Poly A NFT	Aerobacter E. coli NT	Aerobacter
Groin		Pseudomonas E. coli Poly A NFT	E. coli Poly A NFT	Alk. dispar	Alk. dispar
Glans penis			E. coli Poly A NFT E. coli NT Aerobacter Pseudomonas	Proteus	
Anal	E. coli NT E. coli Poly BNFT	E. coli NT E. coli Poly A NFT Pseudomonas	E. coli NT E. coli Poly B 0124:B17	E. coli NT E. coli Poly B 0124:B17	
Feces	E. coli Poly B 0124:B17 086:B7	Aerobacter Alcaligenes Alk. dispar	E. coli NT Proteus Pseudomonas	E. coli NT E. coli Poly B 086:B7, 0124:B17	E. coli NT

Body Area	Sampling Period				
	6	7	8	9	Extra
Axilla					
Groin					
Gland penis					
Anal	Proteus Alk. dispar				
Feces	E. coli NT				E. coli Poly B 0124:B17

TABLE 7 --- Continued

Subject C

Body Area	Sampling Period				
	1	2	3	4	5
Groin		E. coli NT			
Glans penis					
Anal	E. coli NT	Aerobacter E. coli NT	E. coli NT	E. coli NT	
Feces	E. coli NT Alcaligenes Citrobacter	E. coli NT Klebsiella	E. coli NT Aerobacter		

Body Area	Sampling Period			
	6	7	8	9
Groin				
Glans penis				
Anal				
Feces				

TABLE 7 --- Concluded --- Experiment XI

Subject Number	Body Area	Sampling Period											
		1	2	3	4	5	6	7	8	9	10	11	12
41	Feces	E. coli NT	E. coli NT	E. coli NT Aerobacter	E. coli NT	E. coli NT	E. coli NT	E. coli NT	E. coli NT	E. coli NT	E. coli Poly A NFT; Poly B 0128:B12	E. coli NT	E. coli NT
	Groin (Right)	E. coli NT	E. coli NT							E. coli NT			
	Feces	E. coli NT	E. coli NT	E. coli NT	E. coli NT	NR	E. coli NT (Citrobacter)	E. coli NT	NR	E. coli NT	E. coli NT		E. coli NT Aerobacter
42	Groin (Left)	E. coli NT	E. coli NT	NR	NR	E. coli NT	NR	NR	NR	NR	NR	NR	E. coli NT
	Feces	E. coli NT	E. coli NT	NR	E. coli NT	E. coli NT	E. coli NT	E. coli NT	E. coli NT	NR	NR	NR	E. coli NT
	Feces	E. coli NT Proteus	E. coli NT Proteus	E. coli NT Proteus Aerobacter	E. coli NT Proteus	E. coli NT Proteus	E. coli NT Proteus	E. coli Poly A 026:B6 Citrobacter	E. coli Poly A 026:B6 Proteus Aerobacter	E. coli Poly A 026:B6 E. coli NT	E. coli Poly A 026:B6 Aerobacter	E. coli NT Aerobacter	E. coli Poly A 026:B6 E. coli NT Aerobacter
43	Groin (Left)	Aerobacter	Aerobacter	E. coli NT	NR	NR	NR	NR	NR	NR	NR	NR	NR
	Groin (Right)	Aerobacter	Aerobacter	Aerobacter	NR	NR	Aerobacter	NR	NR	Aerobacter	NR	NR	NR
	Gingiva	NR	NR	NR	NR	NR	NR	NR	NR	NR	Pseudomonas	NR	NR

[illegible]

\*formerly *B. anitratum*      NFT = No further type; NT = No type; NR = No recovery

TABLE 8. OCCURRENCE OF CORYNEBACTERIA AND STAPHYLOCOCCI:  
SELECTED BODY AREAS - Experiment X

Subject 37

	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Coryn. Ear Staph.	1						5000 30				0 0
Coryn. Nose Staph.	230 159	1000 34	9320 320	92 47	8800 2810	192 12	569 17	205 64	480 620	289 55	10 480
Coryn. Groin Staph.	2700 1100	>1000 544	0 230	146 36	>8000 2130	4540 2220	> 8000 3600	3170 470	>5000 4650	2370 640	3860 2350
Coryn. G.P. Staph.	4500 250	2440 1970	0 20	0 500	155 334	5500 3500	4550 3320	256 13	60 2	1000 20	740 180
Coryn. Axilla Staph.	0 3	0 2	0 7	0 103	10 750	110 1950	153 660	130 560	10 700	0 230	11 52
Coryn. Toes Staph.	25000 16200	300 25100	3000 20200	520 150	NS	NS	NS	2100 1540	940 680	5300 3000	52400 28800

NS = No sample; subject in evaluator  
Data x 10<sup>4</sup> = total bacteria/gram

TABLE 8 --- Continued --- Experiment X

Subject 38

	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Coryn. Ear Staph.	750						> 6000				600
							5				
Coryn. Nose Staph.	0 120	13 31	0 18	0 4	5 44	8 13	17 17	117 21	126 48	15 4	18 12
Coryn. Groin Staph.	1000 680	2210 50	> 10000 540		4070 1060	11000 980	6600 100	7150 810	1720 800	8400 2000	16200 1500
Coryn. G. P. Staph.	85 12	138 10	95 3	0 0	2350 20	690 150	1610 100	970 190	220 470	6340 150	1710 60
Coryn. Axilla Staph.	0 1	0 4	0 6	0 0	0 3	0 11	0 6	0 53	0 27	0 0	15 13
Coryn. Toes Staph.	0 4300	4500 3200	0 770	0 0	NS	NS	NS	39500 6100	2800 1200	8400 4600	22200 8000

NS = No sample; subject in evaluator

Data x  $10^4$  = total bacteria/gram

TABLE 8 ---- Continued ---- Experiment X

Subject 39

	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Coryn. Ear Staph.	1850 710						672 145				500 170
Coryn. Nose Staph.	60 4	33 2	7 10	106 35	24 16	1300 130	210 40	89 13	141 88	450 67	1630 800
Coryn. Groin Staph.	1000 100	0 420	560 900	9 39	1980 1530	3700 500	> 10000 1170	17700 11300	2470 3100	20200 3800	27500 1800
Coryn. G.P. Staph.	250 61	4000 70	210 300	0 1	49 172	970 430	850 100	364 49	1410 370	1800 110	> 1000 58
Coryn. Axilla Staph.	46 25	TNTC 940	147 21	5 3	123 119	880 2260	5000 4500	3820 380	790 1660	650 650	680 450
Coryn. Toes Staph.	7600 3800	TNTC TNTC	1180 1110	3000 3500	NS NS	NS NS	NS	1240 110	400 900	4100 4700	11300 3900

TNTC = Too numerous to count

NS = No sample; subject in evaluator

Data x 10<sup>4</sup> = total bacteria/gram

TABLE 8 ---- Continued ---- Experiment X

Subject 40

	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Coryn. Ear Staph.	0 2						652 0				4000 0
Coryn. Nose Staph.	82 41	28 38	20 136	2 26	2 14	1900 21	140 10	0 41	112 166	0 5	850 420
Coryn. Groin Staph.	700 730	1050 340	1730 670	0 31	561 1440	4200 3270	200 1300	2090 550	4000 24400	1110 230	3700 1540
Coryn. G.P. Staph.	0 2	0 0	5 5		20 32	27 40	17 48	0 2	6 15	40 14	317 18
Coryn. Axilla Staph.	0 1	0 135	0 2080	0 7	3 103	0 224	0 158	1 19	0 9	0 30	154
Coryn. Toes Staph.	0 800	0 7800	470 5600	1600 8600	NS	NS	NS	540 1150	1400 1400	310 290	1700 2300

NS = No sample; subject in evaluator

Data x  $10^4$  = total bacteria/gram

TABLE 8 ---- Continued ---- Experiment Xa

Subject A

Body Area		Sampling Period								
		1	2	3	4	5	6	7	8	9
Nose	Coryn.	0	35	>300	3	0	0	0	3400	6040
	Staph.	0	20	0	1	1	17	11	10	10
Axilla	Coryn.	0	2	1540	1610	270	60	0	0	0
	Staph.	6	>250	5680	1660	860	410	260	1840	720
Groin	Coryn.	0	0	5000	750	0	3220	1670	2180	3800
	Staph.	2	>375	1220	30	1770	2920	1290	1480	5500
Toes	Coryn.	60	>2000	Suited						
	Staph	70	>3250							

Data x 10<sup>4</sup> = total bacteria/gram



TABLE 8 ---- Continued ---- Experiment Xa

Subject B

Body Area		Sampling Period								
		1	2	3	4	5	6	7	8	9
Nose	Coryn.	1760	0	560	4100	3400	3076	3500	0	14
	Staph	20	5	60	1610	160	630	260	140	8
Axilla	Coryn.	0	0	0	0	6000	460	0	0	0
	Staph.	7	1160		170	9400	610	1600	700	5800
Groin	Coryn.	202	600	1370	3450	5300	1120	4500	8000	6390
	Staph.	5	>2500	120	150	0	280	460	1750	760
Toes	Coryn.	590	710	Suited						
	Staph.	540	2100							
								1180	0	1930
								550	830	2380

Data x  $10^4$  = total bacteria/gram

TABLE 8 ---- Continued ---- Experiment Xa

Subject C

Body Area		Sampling Period								
		1	2	3	4	5	6	7	8	9
Nose	Coryn.	2680	170	164	550	44	153	350	700	1630
	Staph.	0	50	14	190	17	55	94	80	10
Axilla	Coryn.	0	0	0	270	10	0	10	0	0
	Staph.	53	2820	2470	2120	80	230	700	99	480
Groin	Coryn.	>7000	1110	740	1450	>5000	950	1280	2000	7000
	Staph.	40	>2500	40	80	350	150	1180	5000	>5000
Toes	Coryn.	3510	TNTC	Suited						0
	Staph.	540	TNTC							>7000

Data x  $10^4$  = total bacteria/gram

TABLE 8 --- Continued --- Experiment XI

Subject 41

		Sampling Period												
		1	2	3	4	5	6	7	8	9	10	11	12	13
Groin (Left)	Coryn.	960	9560	2850	740	2840	1110	1130	2870	2810	840	710	4060	3320
	Staph.	100	230	480	10	70	10	50	80	90	10	40	220	20
Groin (Right)	Coryn.	2900	-	-	5490	7000	5500	5000	16000	32700	1700	1340	4790	1420
	Staph.	370	-	-	0	2000	20	60	270	700	0	40	80	220

		14	15	16	17	18	19	20	21	22	23	24	25	26
Groin (Left)	Coryn. Staph.	1890	1000	1080	864	925	2090	450	2210	1630	1940	1070	780	1440
		100	247	60	1	173	10	70	740	320	330	900	290	400
Groin (Right)	Coryn. Staph.	320	426	770	1576	1056	2810	1060	1600	560	510	1490	820	1650
		30	157	30	123	77	10	3	270	110	140	300	270	140

- = Confluent growth

Data x  $10^4$  = total bacteria/gram

TABLE 8 ---- Continued ---- Experiment XI

Subject 42

	Sampling Period												
	1	2	3	4	5	6	7	8	9	10	11	12	13
Groin (Left)	Coryn. 620	1470	1280	300	800	2000	200	7200	4800	20	829	1600	150
	Staph. 30	590	30	10	74	105	0	800	130	20	48	207	0
Groin (Right)	Coryn. 1670	-	630	120	531	800	780	10	1068	1228	1240	10030	4110
	Staph. 50	-	0	20	36	96	100	0	92	86	50	230	50

	14	15	16	17	18	19	20	21	22	23	24	25	26
Groin (Left)	Coryn. 858	959	3100	1095	1017	564	268	272	681	681	1100	133	243
	Staph. 29	44	30	84	105	14	22	37	231	149	300	129	200
Groin (Right)	Coryn. 842	1092	1010	898	1150	876	52	233	883	92	900	318	179
	Staph. 26	109	60	46	135	53	29	14	223	175	250	82	150

- = Confluent growth

Data x  $10^4$  = total bacteria/gram

TABLE 8 ---- Continued ---- Experiment XI

Subject 43

	Sampling Period												
	1	2	3	4	5	6	7	8	9	10	11	12	13
Coryn. Groin (Left)	2220	TNTC	TNTC	8600	13400	10000	49200	5320	1300	1340	940	1340	6680
Staph.	20	TNTC	0	100	420	80	800	240	0	880	0	10	0
Coryn. Groin (Right)	1240	8070	TNTC	700	2100	11600	56000	4800	5200	4860	870	4280	1090
Staph.	20	730	3280	300	70	220	600	100	50	300	0	390	0

	14	15	16	17	18	19	20	21	22	23	24	25	26
Coryn. Groin (Left)	4630	3200	2700	8740	7070	3560	450	3030	12280	8980	1740	10	4490
Staph.	30	0	20	0	50	0	0	0	170	110	0	40	140
Coryn. Groin (Right)	900	2410	700	3070	7130	5940	240	1070	10340	6810	2200	0	4080
Staph.	20	0	0	40	90	50	0	50	210	50	0	100	210

- = Confluent growth

Data x 10<sup>4</sup> = total bacteria/gram

TABLE 8 ---- Concluded ---- Experiment XI

Subject 44

	Sampling Period												
	1	2	3	4	5	6	7	8	9	10	11	12	13
Groin Coryn. (Left) Staph.	290	97	6300	1100	2620	4910	1500	55000	42000	30400	16400	58900	21400
	30	60	1370	400	670	790	400	9200	7000	3000	1200	1800	5400
Groin Coryn. (Right) Staph.	620	163	12760	3200	7500	3590	1400	17700	18000	21400	27300	58800	16400
	80	16	1840	0	210	41	600	1700	8000	3400	1200	1600	2800

	14	15	16	17	18	19	20	21	22	23	24	25	25
Groin Coryn. (Left) Staph.	35200	20000	40800	2610	1780	5260	530	1040	550	1790	10800	4100	2500
	3100	1700	5300	930	710	800	140	1220	560	1600	50000	1100	2310
Groin Coryn. (Right) Staph.	12800	19700	3400	610	710	2240	440	1950	6740	1010	7900	11500	2340
	500	600	2200	70	170	130	40	250	160	1120	9300	18400	1140

- = Confluent growth  
 Data x 10<sup>4</sup> = total bacteria/gram

TABLE 9. OCCURRENCE OF CORYNEBACTERIA

Subject 37

Experiment X

Body Area	striatum	pseudodip- theriticum	Pattern				Counts**	
			A	A1	B	B1	Aerobic	Anaerobic
Scalp								
Eye			3				1	
Ear								
Nose		1-5, 10	2, 6, 10*				8	1
Throat								
Gingiva								
Tongue								
Axilla	9		3*, 10*				1	2
Forearm		1*						1
Umbilicus	3		1*, 3				2	1
Groin	1, 1*, 2, 2*, 3*, 6	9	4-6, 8-11 10*	3*, 7	11*		12	6
Glans penis	1*, 2, 6, 8, 10	10, 11*	8-11, 9*	2*, 9	8*	1, 5	12	6
Anal area	1-3, 2*, 4-7		5*, 6, 8-11	2*, 3, 6 8-11			18	3
Toes			8-10		1		4	
Feces								
Total	22	10	30	11	3	2	58	20

Note: Numbers indicate sampling period in which organisms occurred

\* Original isolations taken from an anaerobic blood plate

\*\* Counts indicating relationship between aerobic and anaerobic isolations

TABLE 9 --- Continued

Subject 38  
Experiment X

Body Area	striatum	pseudodip- theriticum	Pattern				Counts**	
			A	AI	B	B1	Aerobic	Anaerobic
Scalp								
Eye								
Ear								
Nose	2*	1, 3, 5, 6, 9, 10*, 11*, 11	1, 8				8	3
Throat								
Gingiva								
Tongue								
Axilla			2*, 7*-10*, 9, 11				2	5
Forearm				1, 11*			1	1
Umbilicus			1*					1
Groin	1, 3, 4*, 6, 7, 11	5, 7, 9-11	2, 5, 8, 11	10	2, 10	2*, 6*	17	3
Glans penis	2-5, 2*, 3*, 5*, 10*	5	1, 5, 8, 9, 11*	10		2*, 6*, 6	11	7
Anal area	1, 4*, 9*	5*	2, 7, 8, 8*, 11	4*, 9		1	7	5
Toes	2		1, 2, 8, 9	11*			5	1
Feces								
Total	19	15	30	5	2	6	51	26

Note: Numbers indicate sampling period in which organisms occurred

\* Original isolations taken from an anaerobic blood plate

\*\* Counts indicating relationship between aerobic and anaerobic isolations



TABLE 9 --- Continued

Subject 39  
Experiment X

Body Area	striatum	pseudodip- theriticum	A	A1	B	B1	B2	Aerobic	Anaerobic
Scalp	1*, 3*	1*							3
Eye									
Ear(1a)								1	
Nose	1, 7*	2, 2*, 3, 6-8 7*	9, 10					9	3
Throat									
Gingiva									
Tongue									
Axilla	1, 3*, 5*, 7		4, 5, 8, 11				6	7	2
Forearm			2					1	
Umbilicus		3*	1*, 2*						3
Groin		1, 3, 6, 7*, 8-10	2*, 4, 6, 7*, 8, 10, 11			9		12	3
Glans penis (9a)	1, 3, 7, 8, 8*, 11		8*	6*			2*	5	5
Anal area	1, 1*, 2, 3, 3*, 5*, 6, 7, 8*, 9, 10		4, 8-10	6, 10*				12	5
Toes	3		1, 2, 4, 10 11					6	
Feces									
Total	33	10	27	3		1	2	54	24

Note: Numbers indicate sampling period in which organisms occurred

\* Original isolations taken from an anaerobic blood plate

\*\* Counts indicating relationship between aerobic and anaerobic isolations

(9a) C. xerosis; (1a) possible C. pyogenes. No liquefaction of gelatin after 5 days.

TABLE 9 --- Continued

Subject 40

Experiment X

Body Area	striatum	pseudodip- theriticum	Pattern				Counts**	
			A	A1	B	B1	Aerobic	Anaerobic
Scalp			1*, 2*					2
Eye			3*					1
Ear								
Nose		1, 3, 6, 7, 9, 4*	2, 5, 8*, 11				8	2
Throat		6*						1
Gingiva								
Tongue		1*, 2*						2
Axilla			3*, 6*, 11*					3
Forearm			3				1	
Umbilicus								
Groin	1, 2, 4, 6		1, 4*, 7-11				10	1
Glans penis	1*, 3, 5, 6		2*, 10				4	2
Anal area	1, 3, 4, 5*		2*, 3*, 5, 6*-8*, 8-11 2*, 4, 6*, 9*				9	9
Toes	1*		4, 8, 10, 11 9				5	1
Feces	10		9*, 11* 11*				1	3
Total	14	9	36	6			38	27

Note: Numbers indicate sampling period in which organisms occurred

\* Original isolations taken from an anaerobic blood plate

\*\* Counts indicating relationship between aerobic and anaerobic isolations

TABLE 9 ---- Continued

Subject A  
Experiment Xa

Body Area	striatum	enzymicum	xerosis	pseudo- diphtheriticum	Patterns					sp.	Acnes
					(A)	A	A <sup>1</sup>	B	B <sup>1</sup>		
Eye						2					
Ear											
Nose				2		3					
Throat		2									
Axilla						5, 9					
Umbilicus											
Groin					3						
Anal area						5					
Feces											
Scalp											
Forearm											
Glans penis											
Toes					2, 7						

Numbers refer to sampling period organisms were isolated

TABLE 9 ---- Continued

Subject B  
Experiment Xa

Body Area	striatum	enzymicum	xerosis	pseudo- diphtheriticum	Patterns					sp.	Acnes
					(A)	A	A <sup>1</sup>	B	B <sup>1</sup>		
Eye											
Ear						1					1
Nose				2, 4, 5, 6, 8				1	3, 7, 9		
Throat										4, 5, 6, 7, 8, 9	
Axilla	5, 6				6	5					
Umbilicus											
Groin	2, 4, 6, 7				1, 3, 4				6	5, 6	
Anal area	1, 3, 5, 6, 7, 8				3, 5	9	4			4	
Feces	3										
Scalp										2	
Forearm	2										
Glans penis	2, 3, 5, 6, 8, 9		6		1	1, 2, 5				9	
Toes						1, 2, 8					

Numbers refer to sampling period organisms were isolated

TABLE 9 ---- Concluded

Subject C  
Experiment Xa

Body Area	striatum	enzymicum	xerosis	pseudo- diphtheriticum	Patterns					sp.	Acnes
					(A)	A	A <sup>1</sup>	B	B <sup>1</sup>		
Eye											
Ear											
Nose				3, 4, 5, 6, 8	1			3, 6	2, 3, 7, 8, 9		
Throat	4									3, 4, 5, 7, 9	
Axilla						1, 4, 5	9				5
Umbilicus	6									4	
Groin	1, 3, 6				3, 4, 5, 7	2, 3, 6, 8					
Anal area	3, 4, 7		4, 9		1, 3, 6	4	2	1		2, 5, 7, 9	
Feces	2					2					
Scalp											
Forearm											
Glans penis	1, 2, 5, 7, 9					4, 5					
Toes					1		7, 9				

Numbers refer to sampling period organisms were isolated

TABLE 10. BIOCHEMICAL REACTIONS OF CORYNEBACTERIA PATTERNS

Pattern	Litmus Milk	Gelatin	Starch	Nitrates	Glucose	Sucrose	Loeffler's	Nutrient Agar	Tellurite	Morphology
A	no change	growth negative no liquefaction	growth no acid	-	-	-	pinpoint to small colony almost translucent at the top of the slant but opaque and cream colored in the heavy growth areas	small grey-white slightly opaque	grey-black colonies	pinpoint almost translucent to small grey slightly opaque
A1*	no change	growth negative	growth negative	-	-	-	small raised cream	white-grey opaque	grey-black colonies	larger colonies opaque
(A)		growth no liquefaction		-	-	-				
B	negative	growth negative	growth $\pm$ acid	$\pm$	acid	acid	small raised glisening slightly translucent at top but cream and opaque at bottom	small colony grey-white slightly opaque	black colonies irregular clumps	grey opaque
B1**	negative	growth negative	growth $\pm$ acid	+	acid	acid				
B2	ARC*** with proteolysis	growth negative	growth $\pm$	+	acid	acid	small cream	medium grey-white slightly opaque	black	grey opaque

\* A1 almost identical to A except in colonial morphology

\*\* B1 probably identical with B except acid is produced in sucrose

\*\*\* ARC - acid reduced curd

TABLE 11. CHROMOGENIC COLONY RECOVERY FROM ACTINO PLATES

Area Sampled	Mycococcus sp.	Mycococcus citreus	Mycococcus albus subspecies lacticus	Proactinomyces sp.	Proactinomyces albus	Proactinomyces citreus	Proactinomyces flavus	Proactinomyces mesentericus	Actinomyces sp.	Actinomyces albus	Actinomyces albus sterilis	Actinomyces albiflavus
Subject 37			Throat (7)		G.P. (8) Ear (3)	Throat (7)						
Subject 38					Ear (3)		Ear (3)	Ear (3)		Ear (3)		
Subject 39						Nose (7)						
Subject 40				Nose (11)								
Subject A											Feces (6)	
Subject B				Feces (6)								
Subject 41									Gingiva* (2, 3)			
Subject 42									Groin* (3, 7, 8, 12)			
Subject 44									Groin (2)*			
Aft Table		X-6										
Bed				Xa-7	X-6, Xa-7					Xa-7		
Floor Psnl. Hyg. Area	Xa-7	X-6		Xa-7	X-6, X-8					Xa-7		
Table					X-8							X-8

G. P. = glans penis

Numbers in parentheses indicate sampling period - Experiment X

\* Experiment XI

TABLE 12. OCCURRENCE OF GRAM-POSITIVE RODS

Subject 37

Experiment X

Sampling Period	Lacto-bacillus	Bacil-laceae	Corynebacterium							
			striatum	pseudodip-theriticum	Pattern					
					A	A1	B	B1	B2	
1	feces		groin, gp, anal	nose, forearm	umbilicus		toe	g. p.		
2			groin, gp, anal	nose	nose	anal, gp				
3			gr, anal	nose	nose, ax	gr, anal				
4	feces		gp, anal	nose	groin					
5	feces	anal	nose	gr, anal				g. p.		
6	throat, gingival feces	g. p.	groin, g. p. anal		nose groin, anal	anal				
7	feces		anal			groin				
8			g. p.		groin, gp, toe, anal	anal	g. p.			
9	feces		axilla	groin	groin, gp, toe, anal	anal g. p.				
10	throat feces		g. p.	nose, gp	nose, gr, axilla, gp, toe, anal	anal				
11	feces		umbil	g. p.	eye, g. p. gr, umbil	anal	groin			



TABLE 12 --- Continued

Subject 38

Experiment X

Sampling Period	Lacto-bacillus	Bacil-laceae	Corynebacterium						
			striatum	pseudodip-theriticum	Pattern				
					A	A1	B	B1	B2
1	feces		anal, gr	nose	umbil. gp, nose forearm			anal	
2	feces		nose, gp, toe		axil, gr, toe, anal		groin	groin, g. p.	
3			groin, gp	nose					
4	throat, ging, fec		anal, gr, gp			anal			
5	feces	g. p.	g. p.	nose, gp, gr, anal	groin, gp				
6	throat		groin	nose				gr, gp	
7	throat, feces			groin	anal, axilla				
8	throat, feces				axilla, gr, anal, gp, nose				
9	throat		anal	nose, gr	axilla g. p. , toe	anal			
10	gingival feces		g. p.	nose, gr	axilla	gr, gp	groin		
11	throat feces		groin	nose, gr	forearm axilla, gr, anal, gp	toe			

TABLE 12--- Continued

Subject 39

Experiment X

Corynebacterium										
Sampling Period	Lacto bacillus	Bacil-laceae	pseudodip-theriticum	Pattern					Miscel-laneous	
				striatum	A	A1	B	B1		B2
1	throat, feces		scalp	nose, gp axil, anal gr, scalp	umbilicus toe					ear*
2	throat, feces		nose	anal	gr, toe				g. p.	
3	throat, feces		nose	axil, anal, gr, gp, toe						
4	throat				axil, gr, toe, anal					
5	throat, feces	anal		axilla, anal	axilla					
6	throat		nose	gr, anal	groin	anal, gp			axilla	
7	throat		nose	nose, anal gr, gp, axilla	umbilicus groin forearm					
8			nose	gr, gp, anal	axil, anal gr, gp					
9	throat			anal, gr	nose, anal			groin		g. p. **
10	feces			gr, anal	nose, toe, gr, anal	anal				
11	feces		nose, umbilicus	gp, scalp	axilla, gr, toe					

\* Possible C. pyogenes

\*\* C. xerosis

TABLE 12--- Continued

Subject 40

Experiment X

Corynebacterium									
Sampling Period	Lacto-bacillus	Bacil-laceae	striatum	pseudodip-theriticum	Pattern				
					A	A1	B	B1	B2
1	feces		gr, anal gp, toe	nose, tongue	groin, scalp				
2	feces		groin		nose, gp anal	anal			
3			g. p., anal	nose	axilla anal				
4			gr, anal	nose	groin, toe	anal			
5		axilla	anal, gp		nose, anal				
6			gr, gp	nose, throat	axilla, anal	anal			
7				nose, tongue	gr, anal scalp				
8	throat				nose, gr, anal, toe				
9				nose	feces gr, anal	anal, toe			
10			feces		gr, anal gp, toe				
11					eye, gr, nose, toe axil, anal forearm feces	feces			

TABLE 12 ---- Continued

Subject A

Experiment Xa

Sample Period	Lacto-bacillus	Bacil-laceae	Corynebacterium									
			striatum	enzy-micum	xerosis	Psd*	(A)	A	A1	B	B1	sp.
1	feces gingiva	feces										
2	throat		throat			nose	toe	eye				
3	feces throat gingiva						groin	nose				
4	throat											
5	throat gingiva							axilla anal				
6	feces											
7	throat						toe					
8	throat											
9	throat gingiva							axilla				

\*Psd. = Pseudodiphtheriticum

TABLE 12 ---- Continued

Subject B

Experiment Xa

Corynebacterium													
Sample Period	Lacto-bacillus	Bacil-laceae	striatum	enzy-micum	xerosis	Psd*	Ⓐ	A	A1	B	B1	sp.	Acnes
1	throat		anal				groin g.p.	toe ear g.p.		nose			ear
2	feces throat		groin forearm g.p.			nose		toe g.p.				scalp	
3	throat	g.p.	g.p. anal feces				groin anal				nose		
4	throat	g.p.	groin			nose	groin		anal			anal throat	
5	throat		g.p. anal axilla			nose	anal	axilla g.p.				axilla groin throat	
6			axilla groin anal g.p.		g.p.	nose	axilla				groin	groin throat	
7	throat		groin anal								nose	throat	
8	throat		g.p. anal			nose		toe			nose	throat	
9	throat		g.p.				anal				nose	g.p. throat	
Extra												feces	

Extra sample taken before run began

TABLE 12 --- Concluded  
Subject C  
Experiment Xa

Corynebacterium													
Sample Period	Lacto-bacillus	Bacil-laceae	striatum	enzy-micum	xerosis	Psd*	(A)	A	A1	B	B1	sp.	Acnes
1			groin g. p.				anal toe ear	axilla		anal		scalp tongue	
2			g. p. feces					feces groin	groin		nose	anal	
3			anal groin			nose	groin anal	groin		nose	nose	throat	
4			throat anal		anal	nose	groin	groin anal axilla				axilla throat	
5			g. p.			nose	groin	g. p. axilla				anal	axilla
6			groin umbilicus			nose	anal	groin		nose		throat	
7			g. p. anal				groin		toe		nose	throat anal	
8						nose		groin			nose		
9			g. p.		anal				axilla toe		nose	anal throat	

TABLE 13. OCCURRENCE OF AEROBES ON BODY AREAS

Experiment X  
(Neisseria)

Subject	Body Area	Sampling Period										
		1	2	3	4	5	6	7	8	9	10	11
37	Gingiva A		X	X		X	X					
	AN		X	X		X	X		X			
	Throat A		X	X	X	X	X	X		X		
	AN		X	X	X	X	X	X	X	X	X	X
	Tongue A	X						X				X
	AN	X						X				X
38	Gingiva A			X			X					
	AN			X		X		X	X			
	Throat A						X					
	AN						X					
	Tongue A	X										
	AN											
39	Eye A	X										
	AN											
	Gingiva A			X				X				
	AN	X	X	X	X	X		X	X	X	X	X
	Throat A			X								
	AN	X				X						
40	Tongue A											
	AN	X						X				X
	Gingiva A		X			X	X					
	AN			X	X	X	X			X		
	Throat A	X					X	X				
	AN		X			X	X			X		
	Tongue A	X						X				X
	AN	X										X

A = Aerobic

AN = Anaerobic

TABLE 13 --- Continued ---

## Experiment X

Body Area	GAFFKYA				SARCINA			
	Subject				Subject			
	37	38	39	40	37	38	39	40
Scalp*	1							
Throat	1, 4, 5, 7, 8, 11	1, 2, 5-8, 10, 11	2, 5-11	2, 4, 7			2	
Tongue*		1, 7, 11	7, 11	7, 11				
Gingival		2, 10, 11	2, 6, 7	5			5	
Axilla							2, 4	

\* Sampled three times only  
 Numbers represent sampling period in which organisms were isolated.



TABLE 13 ---- Continued ---- Experiment Xa

Subject A

Body Area	Hemophilus	Sarcina	Neisseria			Gaffkya*	Miscellaneous
			pharyngitis	catarrhalis	sp.		
Nose						8	
Tongue					2, 3		
Throat					1, 3		
Gingiva					3, 6, 7		
Axilla						3, 5	3, 4, 5, 6**
Groin						3, 5, 6, 7	
Glans penis						5, 6	
Anal						5, 6, 7	
Toe						7, 8	

\* Large Gram-positive cocci resembling Gaffkya. Recovered on phytone-yeast medium

\*\* Fat Gram-negative rod, pinpoint colony on blood agar, oxidase ±, nitrate -, catalase +.

Numbers refer to sampling period organisms were isolated

TABLE 13 ---- Continued ---- Experiment Xa

## Subject B

Body Area	Hemophilus	Sarcina	Neisseria			Gaffkya	Miscellaneous
			pharyngitis	catarrhalis	sp.		
Nose						6, 7	
Tongue					1, 2, 3		
Throat					5, 6, 7, 8, 9		
Scalp Forearm Umbilicus						2	
Gingiva							
Axilla						3, 5, 6, 8	
Glans penis						5, 6, 7	
Groin						3, 4, 6	
Anal						6, 7	
Toe						7	

Numbers refer to sampling period

TABLE 13 ---- Continued ---- Experiment Xa

Subject C

Body Area	Hemophilus	Sarcina	Neisseria			Gaffkya	Miscellaneous
			pharyngitis	catarrhalis	sp.		
Scalp						2	
Tongue							
Gingiva					5		
Axilla							
Glans penis							3**
Groin						6	
Anal							
Toe						8	

\*\* Fat Gram-negative rod, pinpoint colony on blood agar, oxidase  $\pm$ , nitrate -, catalase +.  
 Numbers refer to sampling period

TABLE 13 --- Continued --- Experiment XI

Sampling Period	Subject 41												Subject 42											
	Groin-Left						Groin-Right						Groin-Left						Groin-Right					
	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus
1	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
2	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
3	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
4	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
5	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
6	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
7	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
8	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
9	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
10	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
11	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
12	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
13	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
14	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
15	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
16	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
17	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
18	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
19	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
20	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
21	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
22	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
23	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
24	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
25	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
26	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

X\* = Actinomyces naeslundii  
X\*\* = Strep. salivarius  
X\*\*\* = Enterococcus  
X<sup>B</sup> = Beta hemolytic

TABLE 13 --- Concluded ---- Experiment XI

Sampling Period	Subject 43												Subject 44											
	Groin-Left						Groin-Right						Groin-Left						Groin-Right					
	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus	Staphylococcus	S. aureus	Corynebacterium	Streptococcus	Gram-Neg. Rod	Bacillus
1	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
2	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
3	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
4	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
5	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
6	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
7	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
8	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
9	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
10	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
11	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
12	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
13	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
14	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
15	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
16	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
17	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
18	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
19	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
20	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
21	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
22	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
23	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
24	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
25	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		
26	X	X	X	X			X	X	X	X			X	X	X	X	X		X	X	X	X		

X\* = Enterococcus  
X<sup>B</sup> = Beta hemolytic

TABLE 14. OCCURRENCE OF STAPHYLOCOCCUS AUREUS PHAGE TYPES

Sampling Period																											
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	
3B/3C															X		X		X	X	X		X	X			X
52/52A/80/81 Complex		X			X		X		X	X	X	X			X	X	X		X	X	X	X	X	X			X
47/53/54/75 Complex	X				X								X	X					X								

TABLE 15. RECOVERY AREA OF PHAGE TYPABLE STAPHYLOCOCCUS AUREUS

Sampling Period									
1	2	5	7	9	10	11	12	13	15
52/52A/80/81	Floor Personal Hygiene	Table	43Gingiva	44 Feces	44Gingiva	43 Bed			44Gingiva
3B/3C							42Gingiva		Table Bed
47/53/54/75	Floor personal Hygiene	Floor personal Hygiene						Table	Floor Personal Hygiene

Sampling Period										
	17	18	19	20	21	22	23	24	25	26
52/52A/80/81	Table 43 Bed 44 Nose	Mike*		43 Bed Mike 43 Nose 44	Table Mike 44 Feces	44 Feces	43 Bed	Mike 41Gingiva	Table 43 Bed Mike	Personal Hygiene Seat 44Gingiva
3B/3C	42 Nose		Floor Personal Hygiene Mike	Bed Personal Hygiene Seat 42 Nose	Floor Personal Hygiene 42Gingiva		Bed	Personal Hygiene Seat 42Gingiva		Floor Personal Hygiene Table Bed
47/53/54/75			Personal Hygiene Seat		Personal Hygiene Seat					

\* Microphone

TABLE 16. FUNGI ISOLATED ON PHYTONE-YEAST EXTRACT AGAR  
Experiment X

Sampling Period	Subject Number			
	37	38	39	40
1	Tongue: Candida sp. Nose: Mucor	Ear: Trichosporon Groin: Candida sp. Toe: Candida sp.	Groin: Candida sp. Toe: Candida sp.	Tongue: Rhodotorula
2	Nose: Mucor Toe: T. rubrum Feces: Rhodotorula	Groin: Aspergillus niger Toe: Candida sp.	Nose: Aspergillus niger Toe: Candida sp. Alternaria Feces: Rhodotorula	Thr: Rhodotorula
3	Thr: Rhodotorula Nose: Alternaria Toe: T. rubrum	Thr: Rhodotorula Toe: Candida sp.	Nose: Penicillium sp. Toe: Candida sp.	Thr: Rhodotorula
4	No samples	No samples	No samples	No samples
5	Feces: Rhodotorula	Anal: Aspergillus GP: Aspergillus niger Groin: Trichosporon		Nose: Aspergillus Thr: Candida albicans
6		Nose: Aspergillus	Nose: Penicillium sp. Groin: Rhodotorula	
7	Tongue: Rhodotorula Groin: Trichosporon Anal: Rhodotorula		Toe: Candida sp.	Scalp: T. tonsurans
8	Nose: Alternaria		Nose: Cladosporium	Thr: Rhodotorula Nose: Cladosporium GP: Trichosporon
9			Nose: Penicillium sp.	Toe: Penicillium sp.
10		Thr: Aspergillus	Toe: Trichosporon	
11	Ax: Aspergillus niger Feces: Rhodotorula Scalp: T. tonsurans	Scalp: Alternaria Ax: Cephalosporium	Nose: Cladosporium Scalp: Cladosporium	Ax: Alternaria Scalp: T. tonsurans



TABLE 16 ---- Continued ---- Experiment Xa

Sampling Period	Subject		
	A	B	C
1		Throat: <i>C. gulliermondi</i>	
2	Tongue: <i>C. albicans</i>	Tongue: <i>C. gulliermondi</i> Throat: <i>C. gulliermondi</i> Toe: <i>Aspergillus</i> Nose: <i>Aspergillus</i>	Tongue: <i>C. albicans</i> Umbilicus: <i>Aspergillus</i> Throat: <i>C. albicans</i> Nose: <i>Aspergillus</i>
3		Anal: <i>C. gulliermondi</i>	Feces: <i>Penicillium</i>
4		Throat: <i>C. gulliermondi</i>	
5			
6	Tongue: <i>Candida</i> sp.		Tongue: <i>C. albicans</i> Throat: <i>C. albicans</i>
7	Glans penis: <i>Candida</i> sp.	Throat: <i>C. gulliermondi</i>	
8	Glans penis: <i>Candida</i> sp.	Throat: <i>C. gulliermondi</i>	Throat: <i>C. albicans</i>
9			

TABLE 16 ---- Continued ---- Experiment XI

Sampling Period	Subject Number			
	41	42	43	44
1	Groin(L) Trichosporon Groin(R) Trichosporon		Groin(R) Penicillium sp.	Feces - C. albicans
2	Groin(L) Trichosporon Groin(R) Trichosporon			Gingiva - Penicillium sp.
3	Groin(L) Trichosporon Groin(R) Trichosporon		Groin(L) Trichosporon	
4	Groin(R) Trichosporon Feces - C. albicans Groin(L) Trichosporon			
5	Groin(L) Trichosporon Groin(R) Trichosporon		Groin(R) A. niger	
6	Groin(L) A. niger Groin(R) Trichosporon Room areas: A. niger Penicillium sp.		Groin(L) Penicillium sp.	Groin(L) Penicillium sp.
7	Groin(R) Trichosporon Groin(L) Trichosporon Feces - Rhodotorula			
8	Groin(R) Trichosporon Groin(L) Trichosporon Floor Personal Hygiene: A. niger			Feces - C. albicans
9	Groin(L) Trichosporon Groin(R) Trichosporon			
10	Groin(L) Trichosporon			
11	Groin(L) Trichosporon Groin(R) Trichosporon			Groin(L) Geotrichum

TABLE 16 ---- Continued ---- Experiment XI

Sampling Period	Subject Number			
	41	42	43	44
12	Groin(L)Trichosporon Groin(R)Trichosporon			
13	Groin(L)Trichosporon Groin(R)Trichosporon Bed: Penicillium sp.			
14	Groin(R)Trichosporon Groin(L)Trichosporon		Feces - Rhodotorula	
15	Groin(L)Trichosporon Groin(R)Trichosporon			
16	Groin(L)Trichosporon Groin(R)Trichosporon			Feces - Rhodotorula
17	Groin(R)Trichosporon Groin(L)Trichosporon			
18	Groin(R)Trichosporon Groin(L)Trichosporon			
19	Groin(R)Trichosporon Bed: A. niger	Groin(R)Cladosporium		
20	Groin(R)Trichosporon Groin(L)Trichosporon Bed: A. niger			
21	Groin(L)Trichosporon Floor Personal Hygiene: Penicillium sp.			
22	Groin(L)Trichosporon		Gingiva-Rhodotorula Groin(R) Penicillium sp.	
23	Groin(R)Trichosporon Penicillium sp. Groin(L)Trichosporon			Gingiva - Candida sp.

TABLE 16 ---- Concluded ---- Experiment XI

Sampling Period	Subject Number			
	41	42	43	44
24	Groin(R) Trichosporon Groin(L) Trichosporon		Groin(L) Aspergillus sp.	
25	Groin(R) Trichosporon		Gingiva - Rhodotorula	Gingiva-Aspergillus sp.
26	Groin(L) Trichosporon Gingiva - Rhodotorula Groin(R) Penicillium sp.	Gingiva-Rhodotorula	Gingiva - Rhodotorula	

(R) = right  
(L) = left

TABLE 17. ANALYSIS OF TOTAL COLONIES RECOVERED FROM MAC CONKEY'S PLATES

## EXPERIMENT X

Subject	Sampling Period	E. coli NT	Aerobacter	Providence	Patterns						Isolates per plate
					A	B	C	D	E	F	
37	4	230									230
37	6	142									142
39	9	48	2	29	1						80
39	10	19		43	6	1	2	4	1	4	80

TABLE 17 ---- Concluded ---- Experiment XI

Subject Number	Sampling Period	Escherichia coli				Aerobacter	Pattern A* (+---++)	Total Number per Plate
		No Type	Poly A 026:B6	Poly B 0125:B15	Saline Positive			
41	2	68	0	0	1	0	0	69
43	5	72	0	0	0	0	0	72
41	16	27	0	34*	0	2	1	64
44	16	16	59	0	0	0	0	75
43	15	4	0	0	0	0	0	4

\* TSI A/A + g + - + +

\*\* 14 of these also typed Poly A - no further type

TABLE 18. PATTERNS FOR ENTEROBACTERIACEAE

Description	Indol	Methyl Red	Voges-Proskauer	Simmon's Citrate	Urease	Nitrate	Motility	H <sub>2</sub> S	TSI	Phenolalanine
Pattern A	+	-	+	+	-	+	+	-	A/A+g	-
Pattern B	+	-	+	+	-	+	+	-	A/A+g	+
Pattern C	+	+	+	+	-	+	+	-	A/A+g	+
Pattern D	+	+	+	+	-	+	+	-	A/A+g	-
Pattern E	+	-	+	+	-	+	-	-	A/A+g	-
Pattern F	+	+	-	+	-	+	+	-	A/A+g	-

TABLE 19. MORPHOLOGICAL IDENTIFICATION OF AEROBIC BROTH CULTURES  
Room Areas - Experiment XI

Sampling Period	Date	Microphone Mouthpiece	Personal Hygiene Seat
1	2/28	A B C G	
2	3/1	S B	A B C
3	3/2	S A	A B C
4	3/7	S B A	A B G P
5	3/8	S B A	A C S
6	3/9		
7	3/14	A B	A B C
8	3/15		
9	3/16	C B A	A B C
10	3/21	R A B S	A B C
11	3/22	D A B	A B D R
12	3/23	B S	A B C
13	3/28	A C	
14	3/29	A B S	A B
15	3/30	S B	
16	4/4	A B S	A B
17	4/5	A B	A B
18	4/6	S	A B
19	4/11	S B	A B
20	4/12	A B S	A B
21	4/13		
22	4/18	A S B	A R B
23	4/19	S	B D S
24	4/20	S B	A B
25	4/25	S B	A B G
26	4/26	B G S	A B C

(No data for Sampling Period 8 and 21)



TABLE 19 --- Continued --- Gingiva

Sampling Period	Dilution*	Subject Number			
		41	42	43	44
1	1	S	S	S B	S
	2	S	S	S	S
	3	S	S	S B	S
2	1	S	S A	S	S
	2	S	S	S B	S B
	3	S	S	S B	S
3	1	S	S A	S	S C
	2	S	S A	S	S A
	3	S	S	S A	S
4	1	S	S	S	S
	2	S G	S	S	S
	3	n. o. s.	S A	S	S
5	1	S	S A	S A	S
	2	S	S A B	S A G	S
	3	S A	S C	S A	n. o. s.
6	1		S A	S B	S
	2		S C	S A	S
	3		S C	S	n. o. s.
7	1	S	S A C	A	S
	2	S	S A C G	n. o. s.	S A
	3	S A		n. o. s.	S
8					
9	1	S	S	S	S
	2	G	n. o. s.	n. o. s.	n. o. s.
	3	n. o. s.	n. o. s.	n. o. s.	n. o. s.
10	1	S	S A	S	S A
	2	S	S C	S	S
	3	S G C	S C	C	S C
11	1	S A	S	S	S B
	2	S A	S	S	S G
	3	S	S	n. o. s.	S
12	1	S	S	S	S B
	2	S	S	S	S
	3	S	S	S	S G
13	1			S B	S
	2			S	S B
	3			S	S G

\*#1 =  $10^3$ #2 =  $10^4$ #3 =  $10^5$

TABLE 19 --- Continued --- Gingiva

Sampling Period	Dilution	Subject Number			
		41	42	43	44
14	1	S	S	S	S
	2	S A	R S	S B	S
	3	A B	n. o. s.	S	S
15	1	S	S	S B	S
	2	S	S	S	S
	3	S	S	S	S
16	1	S B	S	S	S
	2	S B	S	S	S
	3	S B	S	S B	S
17	1	S B G	S B	S	S
	2	S G	S	S	S
	3	S G	S	S	S
18	1	S	S	S	S
	2	S	S	S	S
	3	S	S	S	S
19	1	S	S	S B	S
	2	S	S	S B	S
	3	S	S	S B	S
20	1	S	S B	S	S A
	2	S B	S B	S	S A
	3	S B	S B	S	S A
21	1	S A	S	S	S A
	2	S	S	S	S A
	3	S	S	S	n. o. s.
22	1	S B	S B	S	S
	2	S	S	S	S
	3	S	n. o. s.	S	S
23	1	S	S B	S	S
	2	n. o. s.	n. o. s.	n. o. s.	n. o. s.
	3	n. o. s.	n. o. s.	n. o. s.	n. o. s.
24	1	S B	S B	S B	S
	2	S	S B	S G	S
	3	S B	S	S	S
25	1	S	S	S B	S A
	2	S	S	S B	S A
	3	S	S	S B G	A
26	1	S B	S	S B	S
	2	S	S	S B	S
	3	S B	S B	S	n. o. s.

TABLE 19 ---- Continued ---- Groin

Sampling Period	Dilution	Subject Number											
		41			42			43			44		
		Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right
1	1 2 3	C n. o. s. C	A C C C	A B C A B C A B	A B C A B C A B C	A C A C C	A B C A B C A C	A B C A B C A B C	A B C A B C A C	A B C A B C A B C	A C D A C A C	A B C A B C A B C	A C D A C A C
2	1 2 3	A B A B C A B	A B C A B C A B C	B n. o. s. A B C	A B C n. o. s. n. o. s.	A B C A B C A B C	A B C A B C A B	A B C A B C A B C	A B C A B C A B	A B C D n. o. s. n. o. s.	A D A C n. o. s.	A B C D n. o. s. n. o. s.	A D A C n. o. s.
3	1 2 3	A B C A B C A C	A C A B C A C	A C A C A C	A B C A B A B C	A C A C A B C	A B C A B A B C	A C A C A B C	A B C A B C A B C	A B C A B C A B C	A C D A C D A C	A B C A B C A B C	A C D A C D A C
4	1 2 3	A B C A B C C	A C A C B C	A B A B C A B C	A B C A B C B C	A B C A B C A B C	A B C A B C B C	A B C A B C A B C	A B C R A B C R A B C R	A C A B C D	A B C D A C D A C D	A B C A B C A B C	A B C D A C D A C D
5	1 2 3	A C A C A C	A C A C C	A C A C C	A B C A B C B	A B C A B C A B	A B C A B C A B C	A B C A B C A B	A B C A B C A B C	A C A C A C	A B C D A B C A B	A C A C A C	A B C D A B C A B
6	1 2 3	A C A C A C	A C A C B	A B C A B C A B C	A B C A B C A B C	A B C A B C A B	A B C A B C A B C	A B C A B C A B	A B C A B C A B C	A C A C A C	A C A C A C	A C A C A C	A C A C A C
7	1 2 3	A B A B A C	A B C A B C D	A B C A B C B	A C A C A C	A B C A B A B	A B C A B C A B	A B C A B A B	A B C A B C A B	A C A C A C	A C D A C D A C	A C A C A C	A C D A C D A C
8		No Data											
9	1 2 3	A B A B A B	A C D A C A C D	A B C A B C A B C	A B C A B C A B C	A B C A B C A B C	A B C A B C A B C	A B C A B C A B C	A B C A B C A B C	A B C D A B C D A B C	A C D A C D A C D	A B C D A B C D A B C	A C D A C D A C D
10	1 2 3	A C A B C A B	A C A B C C	B C B C C	A B C A B C A C	A B C A B A B	A B C A B A B	A B C A B A B	A B C A B A B	A C D A C D A C	A B C D A B C D C D	A C D A C D A C	A B C D A B C D C D

TABLE 19 --- Continued --- Groin

Sampling Period	Dilution	Subject Number									
		41			42			43			44
		Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right
11	1	AC	ABC	ABC	AB	ABC	AB	ACD	AB	ACD	ABD
	2	AC	ABC	AC	AB	AB	AB	ACD	AB	ACD	ABC
	3	A	ABC	AC	B	AB	AB	AC	AB	AC	ABC
12	1	ABC	ABC	ABC	ABC	ABC	AB	ABDR	AB	ABDR	ABCD
	2	ABC	ABC	AB	ABCS	AB	AB	ABCD	AB	ABCD	ABC
	3	ABC	ABC	AC	ABC	BC	AB	ABC	AB	ABC	BC
13	1	AB	ABC	NO SLIDE			NO SLIDE			NO SLIDE	
	2	AB	ABC								
	3	AB	ABC								
14	1	ABC	ABC	NO SLIDE			NO SLIDE			ACD	ABC
	2	AC	AB							AD	ABC
	3	AB	B							A	ABC
15	1	ABC	ABC	A	ABC	AB	ABC	ABR	ABC	AC	AC
	2	ABC	AB	A	ABC	AB	B	AB	AC	AC	AC
	3	AB	AB	ABC	AB	AB	B	AC	A	A	A
16	1	AC	AB				ABC	ACD	ACD	ACD	ACD
	2	AC	ABC				ABC	D	ABC	ABC	ABC
	3	A	AB				AB	C	C	C	C
17	1	ABC	ABCD	ABC	ABS	BS	AB	AD	ACD	ACD	ACD
	2	AB	ABC	ABC	AB	ABS	AB	AD	AC	AC	AC
	3	AB	ABC	C	AB	BS	AB	A	C	C	C
18	1	ABC	AB	ABC	ABC	ABS	B	ACD	ABC	ABC	ABC
	2	ABC	ABC	AB	ABC	AB	AB	AD	ABC	ABC	ABC
	3	A	AB	B	AC	B	B	A	C	C	C
19	1	ABC	AB	ABC	ABC	ABC	AB	CD	ABD	ABD	ABD
	2	AC	ABC	ABC	AB	AB	B	D	AC	AC	AC
	3	BC	B	ABC	n.o.s.	B	B	n.o.s.	A	A	A
20	1	ABC	AB	ABC	ABC	AB	AB	ABC	ABC	ABC	ABC
	2	AC	ABC	AB	AC	AB	AB	AC	BC	BC	BC
	3	AC	B	C	C	B	ABC	C	BC	BC	BC

TABLE 19 ---- Concluded ---- Groin

Sampling Period	Dilution	Subject Number											
		41			42			43			44		
		Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right	Groin Left	Groin Right
21	1	ABC	AB	ABC	ABC	AB	ABC	AB	ABR	ABC	ABC	ABC	ABC
	2	ABC	AB	ABC	ABC	B	ABC	B	B	ABC	AC	ABC	AC
	3	ABC	AB	ABC	n.o.s.	AC	n.o.s.	AB	B	A	BC	BC	BC
22	1	ABC	ABC	ABC	ABC	AB	AB	B	AB	AD	AC	AD	AC
	2	ABC	ABC	ABC	ABC	A	ABS	B	B	AD	ABC	AD	ABC
	3	AC	AC	AC	AC	AC	AC	AB	AB	A	AC	A	AC
23	1	AC	slide	AC	ABC	AB	ABC	B	AB	AD	AD	AD	AD
	2	AC	broken	AC	AB	AB	AB	B	B	CD	AC	CD	AC
	3	A	AB	A	AB	B	AB	B	B	A	A	A	A
24	1	NO SLIDE			NO SLIDE			AB	BS	ABD	ACD	ABD	ACD
	2							AB	BS	ABD	ABC	ABD	ABC
	3							AB	B	CD	AC	CD	AC
25	1	ABC	AB	ABC	A	ABC	A	BC	AB	ABC	ABC	ABC	ABC
	2	ABC	AB	ABC	A	ABC	A	B	n.o.s.	ABC	AB	ABC	AB
	3	AB	B	AC	A	AC	A	B	AB	C	A	A	A
26	1	ABC	ABC	ABC	ABC	ABC	ABC	B	AB	AC	ABC	AC	ABC
	2	ABC	ABC	ABC	ABC	AB	ABC	B	B	A	ABC	A	ABC
	3	B	ABC	AB	ABC	AB	ABC	B	B	AB	AC	AB	AC

A = large gram positive cocci in pairs and tetrads

B = small gram positive cocci in pairs and tetrads

C = Corynebacteria

D = Gram negative rods

G = medium gram positive rods in pairs and short chains

P = short gram positive rods in chains

R = large gram positive rods, blunt and Bacillus-like

S = Streptococci

n.o.s. = no organisms seen

TABLE 20. RECOVERY OF AEROBES FROM FECES - Experiment X

Sampling Period	Subject 37						Subject 38						Subject 39						Subject 40					
	Staphylococcus	Strep. veridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	Strep. veridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	Strep. veridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	Strep. veridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod
1		X	X	X	X			X		X	X			X		X	X			X		X	X	Gram + Rod
2		X		X				X		X	X			X		X	X			X		X	X	Gram + Rod
3		X		X						X				X		X	X	X		X		X		Gram + Rod
4		X		X	X					X	X			X		X				X		X		Gram + Rod
5		X		X	X			X		X	X			X		X	X							Gram + Rod
6		X		X	X		X	X		X		X				X				X		X		Gram + Rod
7		X		X	X						X								X	X		X		Gram + Rod
8				X			X	X		X	X	X		X		X			X	X				Gram + Rod
9		X		X	X			X		X			X	X		X		X		X				Gram + Rod
10				X	X			X		X	X			X		X	X			X		X		Gram + Rod
11		X		X	X			X		X	X			X		X	X		X	X		X		Gram + Rod

TABLE 20 ---- Continued ---- Experiment Xa

Sampling Period	Subject A						Subject B						Subject C					
	Staphylococcus	Strep. viridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	Strep. viridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	Strep. viridans	Enterococcus	Gram - Rod	Lactobacillus	Gram + Rod
1	X			X	X			X		X				X		X		
2		X		X				X		X	X			X		X		X
3				X	X			X		X		X		X		X		
4	X			X						X								
5	X	X		X						X								
6	X	X		X						X								

TABLE 20 ---- Concluded ---- Experiment XI

Sampling Period	SUBJECT 41						SUBJECT 42						SUBJECT 43						SUBJECT 44									
	Staphylococcus	S. aureus	Enterococcus	Strep. viridans	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	S. aureus	Enterococcus	Strep. viridans	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	S. aureus	Enterococcus	Strep. viridans	Gram - Rod	Lactobacillus	Gram + Rod	Staphylococcus	S. aureus	Enterococcus	Strep. viridans	Gram - Rod	Lactobacillus	Gram + Rod
1			X	X	X	X	X				X	X	X	X	X			X <sup>•</sup>	X	X					X <sup>•</sup>	X	X	X
2			X	X	X							X			X			X	X	X					X	X	X	X
3					X	X	X				X <sup>•</sup>	X		X	X						X					X	X	X
4				X	X	X	X	X				X	X	X					X	X	X	X				X	X	X
5					X	X												X <sup>•</sup>	X	X	X	X				X	X	X
6					X							X	X						X	X	X	X				X	X	X
7			X	X	X		X	X			X	X		X		X			X	X	X	X				X	X	X
8					X	X	X	X			X <sup>•</sup>		X	X	X			X	X	X	X	X				X	X	X
9					X	X	X				X	X	X	X		X <sup>*</sup>		X	X	X	X	X				X	X	X
10			X	X	X	X					X <sup>•</sup>	X					X		X							X	X	X
11					X				X			X			X				X		X					X	X	X
12					X	X						X			X				X		X					X	X	X
13					X	X			X			X			X				X							X	X	X
14					X							X							X <sup>*</sup>	X	X					X <sup>*</sup>	X	X
15				X	X	X						X			X				X <sup>*</sup>	X	X					X <sup>*</sup>	X	X
16			X <sup>•</sup>	X	X	X						X			X						X					X	X	X

• anaerobic strep  
\* coagulase positive



TABLE 21. AEROBIC PLATE COUNTS FROM FECES (1.0 ml)

Experiment X

Subject	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
37	300	92	213	46	150	70	1	53	92	44	325
38	244	181	8	2	17	4	39	16	40	39	169
39	83	102	13	6	4	64	3	73	7	1	24
40	160	113	12	6	N.S.	10	3	2	13	20	72

Experiment Xa

Subject	Sampling Period					
	1	2	3	4	5	6
A	1	2	44	10	10	66
B	59	35	4	150	60	6
C	137	28	44	N.S.	N.S.	N.S.

Data represents bacteria present in  $10^{-7}$  grams of feces  
 N.S. = no sample

TABLE 21 ---- Concluded ---- Experiment XI

Subject Number	Sampling Period															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
41		66	10	15	4	(650)	72	4	42	234	(176)	(132)	6	58	5	88
42		55	0	4	1	NS	2	1	0	0	10	6	1	NS	NS	NS
43	0	2	0	0	0	(0)	0	2	1	(0)	(7)	1	2	3	0	NS
44	69	80	110	167	126	(401)	124	32	37	(236)	(4)	38	25	22	8	65

○ While on contingency diet

Data represents bacteria present in  $10^{-7}$  grams of feces

NS = No Sample

TABLE 22. ANAEROBIC GROWTH\*

## EXPERIMENT X

## Throat Cultures

Subject Number	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
37	- 9	- 8	- 6	- 7	- 9	-10	-10	-10	-10	-10	- 9
38	- 8	- 9	- 6	- 9	- 9	-10	- 7	- 8	-10	-10	-10
39	-10	- 7	-10	- 8	- 8	-10	-10	- 8	-10	- 8	- 8
40	- 8	-10	- 9	- 9	- 8	- 8	-10	- 8	-10	- 9	- 6

## Fecal Cultures

Subject Number	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
37	-14	-13	-13	-12	-14	-13	-13	-12	-12	-11	-12
38	-14	-12	-11	-12	-13	-11	-12	-13	-12	-11	-12
39	-11	-12	-11	-12	-13	-12	-12	-12	-12	-11	-12
40	-12	-13	-13	-13	NS	-12	-13	-12	-11	-11	-11

\*Grams/cc expressed as Log<sub>10</sub>

TABLE 22 ---- Continued ---- Experiment Xa

## Fecal Cultures

Subject Number	Sampling Period					
	1	2	3	4	5	6
A	-12	-12	-12	-12	-13	-13
B	-11	-12	-11	-12	-13	-10
C	-12	-12	-12	NS	NS	NS

## Throat Cultures

Subject Number	Sampling Period							
	1	2	3	4	5	6	7	8
A	-8	-10	-9	-8	-8	-9	-7	-9
B	-10	-10	-10	-10	-9	-10	-10	-9
C	-9	-9	-9	-10	-8	-9	-9	-10

\*Grams/cc expressed as Log<sub>10</sub>

TABLE 22 ---- Concluded ---- Experiment XI

## Fecal Cultures\*

Subject Number	Sampling Period															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
41	-11	-11	-11	-12	-11	-11	-12	-11	-12	-12	-12	-12	-12	-12	-12	-12
42	-11	-11	-12	-11	NS	-14	-13	-12	-12	-13	-12	-12	-11			
43	-10	-11	-11	-12	-11	-11	-11	-12	-11	-10	-11	-13	-12	-12	-12	
44	-11	-11	-12	-12	-12	-13	-14	-13	-13	-13	-12	-11	-12	-12	-13	-13

\*Grams/cc expressed as Log<sub>10</sub>  
 NS = no sample

TABLE 23. MICROORGANISMS ISOLATED FROM FOOD SAMPLES  
(representing all space diets)\*

Foods Sampled	Bacillus sp.	Staphylococci		Enterobacteria	Streptococci	Anaerobes	Yeast	Molds
		(1)	(2)					
Orange-pineapple juice	X						X(sapro- phyte)	
Orange juice								
Grape juice								
Grapefruit juice	X							
Orange-grapefruit juice								
Pea soup	X	X	X					X(Penicillium)
Potato soup	X	X		X(achromo- bacter)	X (viridans)			X (Saprophyte)
Mushroom soup		X						
Corn chowder		X		X(aerobacter)				X (Saprophyte)
Cocoa beverage	X	X						
Tea with lemon & sugar								
Banana pudding								
Butterscotch pudding								
Apricot pudding							X(sapro- phy)	
Chocolate pudding	X							
Bacon and eggs						X (FA-8)		
Bacon squares						X (FA-8)		

(1) Mannitol Negative

(2) Mannitol Positive, Coagulase Negative

\*During second experimental period

TABLE 23 ---- Continued

Foods Sampled	Bacillus sp.	Staphylococci		Enterobacteria	Streptococci	Anaerobes	Yeast	Molds
		(1)	(2)					
Beef sandwich (a)	X							
Beef sandwich (b)	X		X					
Beef and gravy	X	X						
Ham and applesauce	X	X						
Beef pot roast	X	X		X(aerobacter)				
Beef and vegetable			X					
Chicken salad	X							
Chicken sandwich	X							
Chicken and gravy	X	X						
Sausage patties	X							
Shrimp cocktail		X						
Salmon salad	X							
Tuna salad	X		X					
Sugar frosted flakes	X	X						
Creamed green beans	X							
Potato salad	X		X	X(aerobacter)	X (viridans)		X(Rhodo- torula)	
Applesauce								
Creamed carrots	X							
Toasted bread cubes		X						
Appricot cubes	X	X						
Strawberry cereal cubes	X							

TABLE 23 ---- Concluded

Foods Sampled	Bacillus sp.	Staphylococci		Enterobacteria	Streptococci	Anaerobes	Yeast	Molds
		(1)	(2)					
Pineapple cubes	X							.
Cinnamon toast	X							
Toast								
Potatochip blocks	X							
Peanutbutter sandwich		X						
Gingerbread bits	X							
Brownies	X							
Fruit cocktail								
Pound cake								
Apple cereal cubes	X	X						
Date-Fruit cake								
Banana cubes	X							
Pineapple fruit cake	X							
Apricot cereal cube		X		X				
All Star cereal								
Strawberry liquid	X							
Raspberry liquid	X							
Cherry liquid	X							
Butterscotch liquid	X							
Vanilla liquid	X		X					
Chocolate liquid	X							



TABLE 24. OBLIGATE ANAEROBES ISOLATED FROM MISCELLANEOUS BODY AREAS - Experiment X

Subject 37

Body Area	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Anal	7		7					UR	UR		
Axilla											
Ear											
Gingiva	V, P	V, P, UR3	P	M	V, 7, 11			V			V, P, UC
Glans penis								11			P
Nose											
Throat	P	V, UC	UC	P	V			P	UR3	V, P	
Tongue											

ob. = obligate

M = Dialister based on morphology

P = Dialister pneumosintes

UC = Unidentified coccus

UR = Unidentified rod

UR1 = H<sub>2</sub>S+; delayed glucose, sucrose and lactose fermentation, ARC in litmus milk, heavy proteolysis and gasUR2 = H<sub>2</sub>S-; remainder of tests as aboveUR3 = H<sub>2</sub>S-; delayed glucose, sucrose, and lactose fermentation; litmus milk unchangedUR4 = H<sub>2</sub>S-; glucose, sucrose, lactose negative; litmus milk unchanged

V

= Veillonella species

3 = Peptococcus asaccharolyticus

5 = Peptococcus prevotii

7 = Peptococcus constellatus

11 = Peptococcus anaerobius

TABLE 24 ---- Continued ---- Experiment X

Subject 38

Body Area	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Anal											
Axilla											
Ear											
Gingiva		V	V	U, P	V, P, 5	V	V	P	V	V	UC
Glans penis				3, 11							
Nose											
Throat	V		V		3, V	V					V
Tongue	V		V								

TABLE 24 ---- Continued ---- Experiment X

Subject 39

Body Area	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Anal	7	7								11	11
Axilla				11							
Ear		11									
Gingiva	V	V		UR4, UC	V			M	M	V	V
Glans penis			V								5, 11, V
Nose											
Throat	V	UR3, V		V	V		M		V		V
Tongue		V									

TABLE 24 ---- Continued ---- Experiment X

Subject 40

Body Area	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Anal											
Axilla											5
Ear											
Gingiva		V		V	V			V	V		
Glans penis											
Nose								5	5		
Throat	UR		V, P, UR4	V	V	V		V			
Tongue											

TABLE 24 ---- Continued ---- Experiment Xa

Subject A

Body Area	Sampling Period								
	1	2	3	4	5	6	7	8	9
Anal				UC, UR					5, 11
Axilla									5
Gingiva	V	V, UC	V, P	V	UC	V, P	V		
Glans penis				V	V	V			
Groin	3, V				UR, 11				
Throat	V, 11	V	V, UR	5, V	V	V		5, V	
Tongue	P	V							

Subject B

Body Area	Sampling Period						
	1	2	3	4	5	6	7
Anal			UC	UC			
Axilla							
Gingiva*							
Glans penis	UC	3, V					
Groin							
Throat			V	V	V	V	V
Tongue							

\* Not sampled

TABLE 24 ---- Continued ---- Experiment Xa

Subject C

Body Area	Sampling Period								
	1	2	3	4	5	6	7	8	9
Anal				UR, 11					
Axilla									
Gingiva		7		V	V				
Glans penis				11					
Groin									
Throat									
Tongue									

TABLE 24 ---- Continued ---- Experiment XI

## GINGIVA

Subject 41

Organism	Sampling Period																									
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
<i>Peptococcus</i> <i>activus</i>																										
<i>aerogenes</i>																										
<i>grigoroffii</i>														X												
<i>prevotii</i>																										
<i>saccharolyticus</i>																										
<i>asaccharolyticus</i>																										
Miscellaneous			GD3 CN1	FA13			GD3 •	FA10 FA12		X		FA16				•	•	•		FN1				•		•
Unkeyed									X										X							

Subject 42

<i>Peptococcus</i> <i>activus</i>																										
<i>aerogenes</i>																										
<i>grigoroffii</i>	X																			X						
<i>prevotii</i>																										
<i>saccharolyticus</i>																										
<i>asaccharolyticus</i>																										
Miscellaneous		FA13	GD6 •	FA12 FN5	FA13	FN1 •	FA13 •					CT2	•		PS1		FN3	FA13 •		•				•	•	•
Unkeyed		X							X					X												

• = *Veillonella* identified morphologically\* = *P. constellatus*

TABLE 24 ---- Continued ---- Experiment XI

## GINGIVA

Subject 43

Organism	Sampling Period																									
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
<i>Peptococcus</i> <i>activus</i>																										
<i>aerogenes</i>																										
<i>grigoroffii</i>																										
<i>prevotii</i>							X					X												X		
<i>saccharolyticus</i>																										
<i>asaccharolyticus</i>																										
Miscellaneous	FA13	FA13	FA5 FA8	•		•	GD7						GD3						PS2 •	•		•				•
Unkeyed				X															X							

Subject 44

<i>Peptococcus</i> <i>activus</i>																										
<i>aerogenes</i>																										
<i>grigoroffii</i>																										
<i>prevotii</i>																										
<i>saccharolyticus</i>																										
<i>asaccharolyticus</i>																										
Miscellaneous	FA13	FA13	FA13 •	FA13	FA8		CN2	•		FN1		GD3						•						FA13	*	
Unkeyed						X	X																			X

• = *Veillonella* identified morphologically  
 \* = *Veillonella* *alcalescens*



TABLE 24 ---- Continued ---- Experiment XI

## GROIN

Subject 41

Organism	Sampling Period																									
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
<i>Peptococcus activus</i>																										
<i>aerogenes</i>	⊗ X	⊗																								
<i>grigoroffii</i>																X										
<i>prevotii</i>							⊗ X	X										⊗	X							
<i>saccharolyticus</i>		⊗															⊗ X									
<i>asaccharolyticus</i>																										
Miscellaneous		GID-3																								
Unkeyed			X																							

Subject 42

<i>Peptococcus activus</i>																										
<i>aerogenes</i>	⊗																									
<i>grigoroffii</i>																										
<i>prevotii</i>	X	X																⊗								
<i>saccharolyticus</i>						⊗	⊗		X																	
<i>asaccharolyticus</i>																										
Miscellaneous							FA16						⊗													
Unkeyed																										

○ Circle indicates results for right groin

TABLE 24---- Concluded ---- Experiment XI

## GROIN

Subject 43

Organism	Sampling Period																									
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26
<i>Peptococcus</i> <i>activus</i>																										
<i>aerogenes</i>		⊗																								
<i>grigoroffii</i>																										
<i>prevotii</i>			⊗																							
<i>saccharolyticus</i>								X	X	⊗			⊗													
<i>asaccharolyticus</i>					X	X	X																			
Miscellaneous FA5 CT3																										
Unkeyed			⊗						⊗													⊗				

Subject 44

<i>Peptococcus</i> <i>activus</i>																										⊗
<i>aerogenes</i>		X																								
<i>grigoroffii</i>																										
<i>prevotii</i>			⊗				⊗		⊗					X					X		⊗					
<i>saccharolyticus</i>		⊗			X			⊗	X			X														
<i>asaccharolyticus</i>																										
Miscellaneous FA12							FA13																			
Unkeyed																										

○ Circle indicates results for right groin

TABLE 25. RECOVERY OF PEPTOCOCCUS - Experiment XI

	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
Peptococcus											
activus											
aerogenes	41G-R 41G-L 42G-R	41G-R 43G-R 44G-L									
grigoroffii	42 Gin										
prevotii	42G-L	42G-L		43G-R 44 G-R			43 Gin 44G-R 41G-L 41G-R	41G-L 44G-R			
saccharolyticus		44G-R 41G-R			44G-L	42G-R	43G-L 42G-R	43G-L 44G-R	43G-R 44G-L 42G-R		
asaccharolyticus					43G-L	43G-L	43G-L				

Peptococcus	12	13	14-15	16	17	18	19	20	21-24	25	26
activus											44G-R
aerogenes											
grigoroffii				41G-L							
prevotii	43 Gin	44G-L 41 Gin				41G-R	44G-L 41G-L	44G-R 42 Gin		43 Gin	
saccharolyticus	43G-R 44 G-L				41G-R 41G-L						
asaccharolyticus											

G-R = Groin - Right  
 G-L = Groin - Left  
 Gin = Gingiva

TABLE 26. SUMMARY OF FECAL ANAEROBES BY SUBJECT

## Experiment X

Anaerobes	Subject Number			
	37	38	39	40
FA-1	1	3		1
FA-2		1		1
FA-3	4	7	7	6
FA-4				
FA-5			1	
FA-6	1		1	
FA-7		2	1	
FA-8	1			
FA-9	2		1	1
FA-10				
FA-11				
FA-12	7	5	1	
FA-13				
FA-14	1	2	6	
FA-15	4	2	1	5
FA-16			1	1
FA-17		1	1	1
FA-18	1			1
GD-1	1	2	4	3
GD-2				2
GD-3	1	1	1	1
GD-4		2	3	2
GD-5	1	1		1
GD-6			2	4
GD-7		2	4	5
Unkeyed	2	1	6	9
TOTAL	27	30	37	50
FN-1				
FN-2				
FN-3				
FN-4				
FN-5				
Unkeyed Lactobacillus Enterococci Miscellaneous		3	2	1
TOTAL	0	3	2	1

TABLE 26 --- Continued --- Experiment Xa

Anaerobes	Subject Number			Total
	A	B	C	
FA-1		1	1	2
FA-2				0
FA-3	2	1		3
FA-4				0
FA-5	1	2	1	4
FA-6				0
FA-7		1		1
FA-8				0
FA-9			2	2
FA-10		2		2
FA-11				0
FA-12		1		1
FA-13				0
FA-14	3	1		4
FA-15	1			1
FA-16				0
FA-17		1		1
FA-18	3			3
GD-1		1		1
GD-2	3		3	6
GD-3				0
GD-4	4			4
GD-5	2			2
GD-6	1			1
GD-7				0
Unkeyed	6*		1	7
TOTAL	26	11	8	45
FN-1				
FN-2				
FN-3				
FN-4				
FN-5				
Unkeyed Lactobacillus Enterococci Miscellaneous		1		1
TOTAL	0	1	0	1

\* 5 Unkeyed; 1 Eubacterium

TABLE 26 ---- Concluded ---- Experiment XI

Anaerobes	Subject Number*				Total
	41	42	43	44	
FA-1	1	1	3	0	5
FA-2	0	1	1	4	6
FA-3	0	0	4	1	5
FA-4	0	2	0	3	5
FA-5	4	5	4	3	16
FA-6	1	2	3	2	8
FA-7	2	0	2	3	7
FA-8	3	0	2	4	9
FA-9	0	0	3	0	3
FA-10	1	0	0	2	3
FA-11	1	0	2	0	3
FA-12	6	1	5	0	12
FA-13	0	0	0	0	0
FA-14	2	1	1	0	4
FA-15	1	0	3	0	4
FA-16	0	0	1	0	1
FA-17	0	0	0	0	0
FA-18	1	2	1	1	5
GD-1	5	2	1	3	11
GD-2	0	5	2	8	15
GD-3	3	3	2	1	9
GD-4	0	1	0	0	1
GD-5	0	1	4	1	6
GD-6	0	2	0	3	5
GD-7	3	4	0	1	8
Unkeyed	3	3	1	3	10
TOTAL	37	36	45	43	161
FN-1	0	0	0	2	2
FN-2	0	0	0	1	1
FN-3	0	0	0	0	0
FN-4	0	0	0	1	1
FN-5	0	0	0	0	0
Peptococcus grigoroffii	5	0	1	0	6
productus	0	0	0	1	1
Clostridium	0	1	1	0	2
PS3	0	1	0	2	3
CN-1	0	0	3	1	4
CN-2	0	0	1	0	1
CT-3	0	2	0	0	2
TOTAL	5	4	6	8	23

TABLE 27. DISTRIBUTION OF ANAEROBES IN FECAL SAMPLES

Experiment X

Subject 37

Anaerobes	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
FA-1										1	
FA-2											
FA-3				1	1	2					
FA-4											
FA-5											
FA-6						1					
FA-7											
FA-8	1										
FA-9							1	1			
FA-10											
FA-11											
FA-12	1	2	1	2				1			
FA-13											
FA-14	1										
FA-15		2	1	1							
FA-16											
FA-17											
FA-18					1						
GD-1				1							
GD-2											
GD-3							1				
GD-4											
GD-5				1							
GD-6											
GD-7											
Unkeyed										1	1
TOTAL	3	4	2	6	2	3	2	2	0	2	1
FN-1											
FN-2											
FN-3											
FN-4											
FN-5											
Unkeyed											
Lactobacillus											
Enterococci											
Miscellaneous											
TOTAL	0	0	0	0	0	0	0	0	0	0	0

TABLE 27 --- Continued --- Experiment X

Subject 38

Anaerobes	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
FA-1		1						2			
FA-2				1							
FA-3		1	2		1	2		1			
FA-4											
FA-5											
FA-6											
FA-7	1	1									
FA-8											
FA-9											
FA-10											
FA-11											
FA-12							3			1	1
FA-13											
FA-14											
FA-15	2										
FA-16											
FA-17										1	
FA-18											
GD-1			2								
GD-2											
GD-3				1							
GD-4					2						
GD-5					1						
GD-6											
GD-7			1							1	
Unkeyed		1									
TOTAL	3	4	5	2	4	2	3	3	0	3	1
FN-1											
FN-2											
FN-3											
FN-4											
FN-5											
Unkeyed											
Lactobacillus											
Enterococci											
Miscellaneous	2*									1**	
TOTAL	2	0	0	0	0	0	0	0	0	1	0

\* PS<sub>2</sub>\*\* PS<sub>3</sub>



TABLE 27 --- Continued --- Experiment X

Subject 39

Anaerobes	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
FA-1											
FA-2											
FA-3			1	2			1		1		2
FA-4										1	
FA-5								1			
FA-6											
FA-7							1				
FA-8											
FA-9					1						
FA-10											
FA-11											
FA-12		1									
FA-13											
FA-14					1			1			
FA-15		1									
FA-16											1
FA-17											1
FA-18											
GD-1			1		1	1		1			
GD-2											
GD-3					1						
GD-4		1				1		1			
GD-5											
GD-6			1							1	
GD-7		2							1	1	
Unkeyed	1								1	3	1
TOTAL	1	5	3	2	4	2	2	4	3	6	5
FN-1											
FN-2											
FN-3											
FN-4											
FN-5											
Unkeyed Lactobacillus Enterococci Miscellaneous	2*										
TOTAL	2	0	0	0	0	0	0	0	0	0	0

\*PS<sub>3</sub>

TABLE 27 --- Continued --- Experiment X

Subject 40

Anaerobes	Sampling Period										
	1	2	3	4	5	6	7	8	9	10	11
FA-1				1							
FA-2		1									
FA-3	1					1	1	1	1		1
FA-4											
FA-5											
FA-6											
FA-7											
FA-8											
FA-9										1	
FA-10											
FA-11											
FA-12											
FA-13											
FA-14		2				2				1	1
FA-15				1		2	1		1		
FA-16										1	
FA-17	1										
FA-18				1							
GD-1			2	1							
GD-2				1					1		
GD-3						1					
GD-4		1								1	
GD-5	1										
GD-6			1			1					2
GD-7		2				1		1		1	
Unkeyed	1	2				2		1		2	1
TOTAL	4	8	3	5	0	10	2	3	3	8	5
FN-1											
FN-2											
FN-3											
FN-4											
FN-5											
Unkeyed Lactobacillus Enterococci Miscellaneous				1*						1	
TOTAL	0	0	0	1	0	0	0	0	0	1	0

\* PS<sub>1</sub>

TABLE 27 --- Continued --- Experiment Xa

Subject A

Anaerobes	Sampling Period						
	1	2	3	4	5	6	Total
FA-1							
FA-2							
FA-3			2				2
FA-4							
FA-5		1					1
FA-6							
FA-7							
FA-8							
FA-9							
FA-10							
FA-11							
FA-12							
FA-13							
FA-14	2				1		3
FA-15		1					1
FA-16							
FA-17							
FA-18		1	1	1			3
GD-1							
GD-2				1	1	1	3
GD-3							
GD-4				1	3		4
GD-5					1	1	2
GD-6				1			1
GD-7							
Unkeyed	2		2	1(a)		1	6
TOTAL	4	3	5	5	5	4	26
FN-1							
FN-2							
FN-3							
FN-4							
FN-5							
Unkeyed							
Lactobacillus							
Enterococci							
Miscellaneous							
TOTAL	0	0	0	0	0	0	0

(a) Eubacterium

TABLE 27 --- Continued --- Experiment Xa

Subject B

Anaerobes	Sampling Period						
	1	2	3	4	5	6	Total
FA-1		1					1
FA-2							
FA-3					1		1
FA-4							
FA-5		1		1			2
FA-6							
FA-7	1						1
FA-8							
FA-9							
FA-10	2						2
FA-11							
FA-12			1				1
FA-13							
FA-14	1						1
FA-15							
FA-16							
FA-17	1						1
FA-18							
GD-1			1				1
GD-2							
GD-3							
GD-4							
GD-5							
GD-6							
GD-7							
Unkeyed							
TOTAL	5	2	2	1	1	0	11
FN-1							
FN-2							
FN-3							
FN-4							
FN-5							
Unkeyed							
Lactobacillus				1			1
Enterococci							
Miscellaneous							
TOTAL	0	0	0	1	0	0	1

TABLE 27 --- Continued --- Experiment Xa

Subject C

Anaerobes	Sampling Period						
	1	2	3	4	5	6	Total
FA-1 FA-2 FA-3			1				1
FA-4 FA-5 FA-6	1						1
FA-7 FA-8 FA-9	2						2
FA-10 FA-11 FA-12							
FA-13 FA-14 FA-15							
FA-16 FA-17 FA-18							
GD-1 GD-2 GD-3 GD-4		1	2				3
GD-5 GD-6 GD-7 Unkeyed	1						1
TOTAL	4	1	3	NS	NS	NS	8
FN-1 FN-2 FN-3 FN-4 FN-5							
Unkeyed Lactobacillus Enterococci Miscellaneous							
TOTAL	0	0	0	NS	NS	NS	0

TABLE 27 --- Continued --- Experiment XI

Subject 41

Anaerobes	Sampling Period																
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
FA-1		Culture Not Transferable												1			
FA-2																	
FA-3																	
FA-4	3												1				
FA-5																1	
FA-6																	
FA-7	1													1			
FA-8				1	1	1											
FA-9																	
FA-10									1						1		
FA-11									1	1	1	1				1	1
FA-12																	
FA-13																	
FA-14													1				1
FA-15												1					
FA-16																	
FA-17																	
FA-18							1										
GD-1	1			2	1												1
GD-2																	
GD-3								1						1			1
GD-4																	
GD-5																	
GD-6																	
GD-7																	
Unkeyed			2	1									1		1	1	
TOTAL	5			4	3	1	3	0	2	1	1	3	3	2	3	2	4
FN-1																	
FN-2																	
FN-3																	
FN-4																	
FN-5																	
FN-5																	
Peptococcus grigoroffii				1	1		2							1			
TOTAL	0		0	1	1	0	2	0	0	0	0	0	0	1	0	0	

TABLE 27 --- Continued --- Experiment XI

Subject 42

Anaerobes	Sampling Period															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
FA-1					Culture Not Transferable							1				
FA-2								1								
FA-3																
FA-4		1				1										
FA-5		1		2								1	1			
FA-6			1										1			
FA-7																
FA-8																
FA-9																
FA-10																
FA-11																
FA-12			1													
FA-13						1										
FA-14																
FA-15																
FA-16																
FA-17												1	1			
FA-18																
GD-1						1				1						
GD-2	1						1			1	1	1				
GD-3			1					1		1						
GD-4									1							
GD-5								1	1			1				
GD-6																
GD-7						1					1	1	1			
Unkeyed			1			1				1						
TOTAL	1	2	4	2		5	1	3	2	4	2	6	4	0	0	0
FN-1					Culture Not Transferable											
FN-2																
FN-3																
FN-4																
FN-5																
PS3	1															
CT3																
Clostridium						1					1	1				
TOTAL	1	0	0	0		1	0	0	0	0	1	1	0	0	0	0

TABLE 27 --- Continued --- Experiment XI

Subject 43

Anaerobes	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
FA-1		Culture Not Transferable	1	1	1											
FA-2												1				
FA-3	1								2						1	
FA-4																
FA-5			1	1			1		1							
FA-6			2										1			
FA-7							1						1			
FA-8						1							1			
FA-9				1				1				1				
FA-10																
FA-11														1	1	
FA-12			2		1					1				1		
FA-13					1											
FA-14																
FA-15										2		1				
FA-16											1					
FA-17																
FA-18						1										
GD-1											1					
GD-2						1		1								
GD-3					1									1		
GD-4																
GD-5					1						2			1		
GD-6																
GD-7																
Unkeyed							1									
TOTAL	1		6	3	5	3	3	2	3	3	4	3	3	4	2	0
FN-1		Culture Not Transferable														
FN-2																
FN-3																
FN-4																
FN-5																
CN-1												2		1		
CN-2						1										
Clostridium *								1					1			
TOTAL	0		0	0	0	1	0	1	0	0	0	2	1	1	0	0

\*Peptococcus grigoroffii



TABLE 27 --- Concluded --- Experiment XI

Subject 44

Anaerobes	Sampling Period															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
FA-1	Culture Not Transferable	Culture Not Transferable			1							2			1	
FA-2																1
FA-3																1
FA-4			1					1	1		2					
FA-5								1	1			1				
FA-6										1		1				
FA-7								1	1	1	1					
FA-8							1			1	1				2	
FA-9																
FA-10									1					1		
FA-11																
FA-12																
FA-13																
FA-14																
FA-15																
FA-16													1			
FA-17																
FA-18																
GD-1	Culture Not Transferable	Culture Not Transferable						2	1	1	1			1	2	2
GD-2														1		
GD-3				1												
GD-4																
GD-5			1													
GD-6														1	2	
GD-7								1								
Unkeyed			1				1				1					
TOTAL			3	1	1	0	2	4	4	3	6	4	1	5	7	3
FN-1															2	
FN-2				1												
FN-3																
FN-4														1		
FN-5																
PS3									1	1						
CN1							1									
*							1									
Unkeyed																
TOTAL	0	0	0	1	0	0	2	0	1	1	0	0	0	1	2	0

\* Peptostreptococcus productus

TABLE 28. SUMMARY OF FECAL ANAEROBES BY SAMPLING PERIOD

## Experiment X

Anaerobes	Sampling Period											
	1	2	3	4	5	6	7	8	9	10	11	Total
FA-1		1		1				2		1		5
FA-2		1		1								2
FA-3	1	1	3	3	2	5	2	2	2		3	24
FA-4												0
FA-5										1		1
FA-6						1		1				2
FA-7	1	1					1					3
FA-8	1											1
FA-9					1		1	1		1		4
FA-10												0
FA-11												0
FA-12	1	3	1	2			3	1		1	1	13
FA-13												0
FA-14	1	2			1	2		1		1	1	9
FA-15	2	3	1	2		2	1		1			12
FA-16										1	1	2
FA-17	1									1	1	3
FA-18				1	1							2
GD-1			5	2	1	1		1				10
GD-2				1					1			2
GD-3				1	1	1	1					4
GD-4		2			2	1		1		1		7
GD-5	1			1	1							3
GD-6			2			1				1	2	6
GD-7		4	1			1		1	1	3		11
Unkeyed	2	3				2		1	1	6	3	18
TOTAL	11	21	13	15	10	17	9	12	6	18	12	144
FN-1												
FN-2												
FN-3												
FN-4												
FN-5												
Unkeyed Lactobacillus Enterococci Miscellaneous	4									2		6
TOTAL	4	0	0	0	0	0	0	0	0	2	0	6

TABLE 28 --- Continued --- Experiment Xa

Anaerobes	Sampling Period						Total
	1	2	3	4	5	6	
FA-1		1	1				2
FA-2		1					1
FA-3			2		1		3
FA-4							0
FA-5	1	2		1			4
FA-6							0
FA-7	1						1
FA-8							0
FA-9	2						2
FA-10	2						2
FA-11							0
FA-12			1				1
FA-13							
FA-14	3				1		4
FA-15		1					1
FA-16							
FA-17	1						1
FA-18		1	1	1			3
GD-1			1				1
GD-2		1	2	1	1	1	6
GD-3							0
GD-4				1	3		4
GD-5					1	1	2
GD-6				1			1
GD-7							0
Unkeyed	3		2	1(a)		1	7
TOTAL	13	7	10	6	6	4	46
FN-1							
FN-2							
FN-3							
FN-4							
FN-5							
Unkeyed Lactobacillus Enterococci Miscellaneous				1			1
TOTAL	0	0	0	1	0	0	1

(a) Eubacterium

TABLE 28 --- Concluded --- Experiment XI

Anaerobes	Sampling Period															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
FA-1			1	1	1							1		1		
FA-2					1			1				3			1	
FA-3	1								2						1	1
FA-4		1	1			1					2					
FA-5	3	1	3	1			1	1	2			3	1			
FA-6			3							1		1	2		1	
FA-7	1						1		1	1	1		2			
FA-8				1	1	2	1				1		1		2	
FA-9				1				1				1				
FA-10									1					2		
FA-11								1						1	1	
FA-12			3		1			1	1	2	1			1	1	1
FA-13																
FA-14					1	1						1				1
FA-15										2	1	1				
FA-16											1					
FA-17																
FA-18						2						1	2			
GD-1	1		2	1		1				1	1			1		3
GD-2	1					1	1	3	1	2	2	1		1	2	
GD-3			1	1	1	1		1		1		1		1		1
GD-4									1							
GD-5			1		1					2		1		1		
GD-6								1	1					1	2	
GD-7						1		1			2	1	2	1		
Unkeyed			4	1		1	2			1	1					
TOTAL	7	2	19	7	7	11	6	11	10	13	13	16	10	11	11	7
FN-1															2	
FN-2				1												
FN-3																
FN-4														1		
FN-5																
P. gregoroffi				1	1		2						1	1		
P. productus							1									
Clostridium								1				1				
PS3	1								1	1						
CN1							1					2		1		
CN2						1										
CT3						1					1					
TOTAL	1	0	0	2	1	2	4	1	1	1	1	3	1	3	2	0

**TABLE 29. ANAEROBIC FECAL ISOLATES ACCORDING TO RANK  
OF OCCURRENCE - COMPARISON OF THREE STUDIES**

<b>Baseline Study NASw-738*</b>	<b>Indigenous Microflora Study AF33(615)-1814**</b>	<b>Current Study AF33(615)-3255***</b>
FA-1	FA-15	FA-3
FA-15	FA-3	FA-12
FA-3	FA-18	GD-1
FA-5	FA-12	GD-7
FA-12	FA-1	GD-2
FA-6	FA-14	FA-5
FA-14	FA-5	FA-15
FA-8	FA-17	FA-14
FA-10	FA-9	GD-3
FA-18	FA-7	GD-6
FA-17	FA-8	FA-1 )
FA-2	FA-6	FA-7 :
FA-16	GD-6	FA-8 :
FA-11	FA-10	FA-6 )
FA-7	GD-3	GD-5
FA-9	GD-1	FA-2
FA-13	FA-2	GD-4
FA-4	FA-16	FA-18 )
	GD-5	FA-9 }
	GD-2 }	FA-4
	GD-7 }	FA-16
	GD-4	FA-17 )
	FA-13 }	FA-10 :
	FA-4 }	FA-11 )
	FA-11	

\* Study of the Normal Fecal Bacterial Flora of Man, L. S. Gall, NASA CR-467, June 1966.

\*\* Determination of the Indigenous Microflora of Men in Controlled Environments, P. E. Riely, D. Geib, D. Shorenstein, AMRL, Wright-Patterson A. F. B. , Ohio.

\*\*\* Research on Microbiological Flora of Human Subjects Undergoing Conditions of Simulated Environment, AMRL, Wright-Patterson A. F. B. , Ohio.

TABLE 30. PRESENTATION OF CONDENSED DATA

Area	Experimental Period	Range	Mean	Median	Mode	Standard Deviation of Mean
Glans Penis	pre-Evaluator	0 - 970	163	48	500	231
	17-18 Day	12 - 1280	354	165	-	387
	22-23 Day	2 - 3750	607	97	-	1074
	25 Day	2 - 6400	1177	263	-	1890
	post-Evaluator	0 - 5100	561	108	650	1098
Groin	pre-Evaluator	0 - 15440	9910	329	500	2288
	11-12 Day	4 - 5760	1719	1000	1000	1923
	17-18 Day	64 - 233000	3098	1000	1000	5273
	22-23 Day	58 - 10000	2488	1000	1000	2830
	25 Day	2 - 16700	3651	1190	1000	4225
	post-Evaluator	0 - 46400	7181	1000	250 & 1000	11791
Axilla	pre-Evaluator	0 - 5030	598	96	250	1131
	11-12 Day	0 - 5160	914	180	-	1513
	14-15 Day	3 - 9000	2287	1550	400	2689
	17-18 Day	6 - 10000	3320	1070	-	3933
	25 Day	4 - 26700	3691	1000	1000	2947
	post-Evaluator	0 - 75000	5470	1000	1000	15211

TABLE 30 ---- Concluded.

Area	Experimental Period	Range	Mean	Median	Mode	Standard Deviation of Mean
Gingiva	pre-Evaluator	0 - 100000	6168	171	-	21691
	25 Day	9 - 3510	523	184	-	1053
	post-Evaluator	2 - 5790	917	307	-	1411
Anal Area	pre-Evaluator	0 - 10000	1483	415	500 & 1000	2322
	11-12 Day	10 - 7700	1525	185	-	2369
	14-15 Day	10 - 28600	7301	7229	-	1017
	17-18 Day	10 - 15000	2083	285	-	4236
	25 Day	14 - 32900	4020	420	-	7682
	post-Evaluator	7 - 40900	4379	500	310 & 1000	8585
Toe	pre-Evaluator	75 - 10000	2356	1000	1000	2886
	25 Day	2 - 40000	7188	1700	2000 & 1000	11628
	post-Evaluator	0 - 92000	18780	1000	1000	27387

**TABLE 31. MORPHOLOGY AND BIOCHEMICAL REACTIONS OF  
FECAL ANAEROBES**

Type Culture	Morphology	Agar Shake	pH Broth*	Growth on Meat Infusion Agar	Gelatin Liquefaction	Litmus Milk	H <sub>2</sub> S	Nitrate Reduction	Indole	Glucose	Lactose	Maltose	Sucrose	Dextrin	Gas Produced in Culture Media	Enriched Culture Media**	Peptone Water
FA-1	sl gr + rods	ob an	7.0 4.6	+	-	R	-	-	-	Alk	Acid	Acid	Acid	Alk	-	-	-
FA-2	sl gr + rod, tadpole	ob an	6.4 4.5	-	-	ARC	-	-	-	Acid	Acid	Acid	Acid	Alk	-	+	-
FA-3	gr neg elongate pt rds in pr	ob an heavy gas	7.5 6.1	+	-	1/2 R	+	-	(+)	Alk	Alk	Alk	Alk	Alk	+	NR	-
FA-4	sl gr + rods	ob an	5.6 4.65	+	-	ARC	-	-	±	Acid	Acid	Acid	Acid	Alk	-	-	-
FA-5	sl med gr + rod clusters	ob an	5.5 4.55	-	-	ARC	-	-	-	Acid	Acid	Acid	Acid	Acid	-	+	-
FA-6	gr + med rod clusters	ob an	6.6 4.45	+	-	ARC	-	-	+	Acid	Acid	Acid	Acid	Alk	-	-	-
FA-7	sm gr neg sl rod bipolar	ob an	6.6 4.85	-	-	ARC	-	-	±	Acid	Acid	Acid	Acid	Alk	-	+	-
FA-8	tiny gr neg sl rods, sl curve	ob an	6.9 8.0	+	-	1/2 R	-	-	-	Alk	Acid	Acid	Acid	Acid	-	-	-
FA-9	pleo gr + rod hooked chains	ob an	7.0 4.85	-	-	1/2 R	-	-	-	Acid	Alk	Acid	Alk	Alk	-	+	-
FA-10	v sm gr + rods in chain bipolar sl pt	ob an	6.7 4.90	+	-	ARC	-	-	-	Acid	Alk	Acid	Alk	Alk	-	-	-
FA-11	sh med gr + rods	ob an	6.5 4.5	+	-	ARC	-	-	-	Acid	Acid	Acid	Acid	Alk	-	-	-
FA-12	tiny pt gr + rods chains coccoid	ob an	7.2 4.65	+	-	1/2 ARC	-	-	-	Acid	Acid	Acid	Acid	Alk	+	-	-
FA-13	sm gr neg cocci in masses	ob an hvy gas	6.7 8.1	-	-	R	+	-	-	Acid	Acid	Acid	Acid	Acid	+	+	+
FA-14	gr neg rods long sl with gr + areas	ob an hvy gas	6.7 5.3	+	-	R	-	-	-	Acid	Alk	Alk	Acid	Alk	+	-	-
FA-15	sh fat gr neg rods pt ends	ob an hvy gas	6.7 4.65	+	-	ARC	+	-	-	Acid	Acid	Acid	Acid	Acid	+	-	+
FA-16	gr + pleo rods tadpole	anaerobic collar	6.8 4.62	-	-	ARC	-	-	-	Acid	Acid	Acid	Acid	Acid	-	+	-
FA-17	lg gr + rod palisades and V's	ob an sl gas	6.6	-	-	ARC	-	-	-	Acid	Alk	Acid	Alk	Alk	±	+	-
FA-18	gr + sl rod irregular staining	ob an	6.3 6.6	+	-	ARC	-	+	-	Acid	Acid	Acid	Acid	Acid	-	-	+
GD-1	sh gr neg rod pairs and chains	ob an heavy gas	6.7 5.4	+	-	ARC	+	-	-	Acid	Alk	Alk	Alk	Alk	-	-	-
GD-2	sh gr neg rod in pairs	ob an	6.2 6.4	+	-	ARC	-	-	-	Acid	Acid	Acid	Acid	Acid	-	-	-
GD-3	gr neg pt rods	ob an	6.8	+	-	R	±	-	-	Alk	Alk	Alk	Alk	Alk	-	-	-
GD-4	gr neg sl rods, pleo	ob an heavy gas	6.3 6.4	+	-	ARC	±	-	+	Acid	Acid	Acid	Acid	Acid	+	-	-
GD-5	gr ± med rods in chains	ob an	6.2 6.6	+	-	ARC	+	-	-	Acid	Acid	Acid	Acid	Acid	-	-	+
GD-6	gr neg rods pleo pairs	ob an heavy gas	5.9	+	-	ARC	-	-	+	Acid	Acid	Acid	Acid	Acid	+	-	-
GD-7	gr ± short pleo rods in pairs	ob an heavy gas	6.8	+	-	R	±	-	-	Acid	Acid	Acid	Acid	Alk	+	-	+

\* Top number pH = 1/10% glucose heavily buffered; Bottom = 5/10% glucose not buffered  
\*\* Serum required



TABLE 32. MORPHOLOGY AND BIOCHEMICAL REACTIONS OF REPRESENTATIVE AMERICAN TYPE CULTURES

Culture	Morphology	Agar Shake	pH Broth	Growth on Meat Infusion Agar	Gelatin Liquefaction	Litmus Milk	H <sub>2</sub> S	Nitrate Reduction	Indol	Glucose	Lactose	Maltose	Sucrose	Gas Produced in Culture Media	Enriched Culture Media Required	Gas in Peptone H <sub>2</sub> O
<i>Clostridium butyricum</i>	Gram + rod, sub-terminal spores	obligately anaerobic; heavy gas	6.2	+	-	ARC proteolysis	-	+	-	A 48	A 48	A 48	A 48	+	-	+
<i>Clostridium centro-sporigenes</i>	Gram + rod, central spores	obligately anaerobic; heavy gas	6.8	+	+	complete digestion	+	-	+	A 24	A 72	A 72	A 24	+	-	no growth
<i>Eubacterium limosum</i>	Gram +, medium, curved rods, some oval, pleomorphic	facultatively anaerobic, slight gas	6.5	+	-	ARC	-	-	+	A 48	A 4d	A 72	sl A 4d	+	-	no growth
<i>Fusobacterium polymorphum</i>	Gram - pleomorphic rods, long & slender, undulating filaments	obligately anaerobic	6.7	+	-	reduced	+	-	+	A 72	A 72	A 72	A 72	-	-	no growth
<i>Lactobacillus bifidus</i>	Gram + rod small slender	obligately anaerobic	7.0	+	-	no growth	-	-	-	-	-	-	A 24	-	-	no growth
<i>Lactobacillus acidophilus</i>	Gram + rod rounded ends in pairs and chains	facultatively anaerobic	6.3	-	-	ARC proteolysis	-	-	-	A 24	A 48	A 72	A 24	-	+	no growth
<i>Propionibacterium acnes</i>	Gram + small rod clubbed	facultatively anaerobic	6.6	+	-	reduced	-	-	+	A 48	sl A 72	A 72	sl A 72	-	-	-
<i>Ranibacterium pseudorammosum</i>	Gram + slender rod some branching	obligately anaerobic	6.7	-	+	ARC	-	-	-	A 48	sl A 72	A 72	sl A 72	-	+	-
<i>Sphaerophorus freudentii</i>	Gram - short oval rod, coccoid and swollen forms	facultatively anaerobic, heavy gas	6.9	+	+	reduced	+	+	-	A 48	A 48	A 48	A 24	+	-	+
<i>Sphaerophorus necrophorus</i>	Gram - short pleomorphic rod, swollen areas	obligately anaerobic	6.3	+	-	delayed ARC	-	-	+	A 4d	A 48	A 4d	A 48	+	-	no growth

TABLE 33. PHYSIOLOGICAL CHARACTERISTICS OF TYPE CULTURES\*

Type Culture	% Lactic Acid/ Wt. Glucose	% Substrate Converted to $\text{NH}_3$	Decarboxylation			
			Lysine	Histidine	Tyrosine	Arginine
Lactic Acid Forming Predominating Fecal Anaerobes	FA-2	26	2	0	0	+
	FA-4	39	2	0	0	0
	FA-5	40	2	0	0	0
	FA-11	37	2	X	0	0
	FA-16	40	2	0	0	+
Deaminating and Decarboxylating Predominating Fecal Anaerobes	FA-1	5	13	0	+	+
	FA-9	26	16	+	+	+
	FA-10	20	12	+	+	+
	FA-12	19	28	+	+	+
	FA-7	28	12	0	+	+
	FA-8	28	23	0	+	0
Miscellaneous Predominating Fecal Anaerobes	FA-3	9	6	+	+	+
	FA-6	9	2	0	0	0
	FA-13	Used	2	(+)	(+)	(+)
	FA-14	9	2	+	+	+
	FA-15	21	9	0	0	+

( ) = Questionable results due to gas formation by culture

X = Test not done

\* = Results obtained under NASA contract NASw-738, "Study of the Normal Fecal Bacterial Flora of Man."  
NASA CR-146.

TABLE 34. BIOCHEMICAL PROPERTIES OF BACTEROIDES, FUSOBACTERIUM AND MOTILE ANAEROBES MOST CLEARLY IDENTIFIED AS INDIGENOUS TO MAN\*

\*"BIOCHEMICAL PROPERTIES OF BACTEROIDES, FUSOBACTERIUM, AND MOTILE ANAEROBES MOST CLEARLY IDENTIFIED AS INDIGENOUS TO MAN"

	Motility	Hemolysis	Capsule	Odor	Gelatin liquefied	Indole formed	H <sub>2</sub> S produced	Nitrate reduced	NH <sub>3</sub> formed	Growth in peptone water	Final pH in glucose	Milk	Gas formed	Glucose	Sucrose	Mannitol	Glycerol	Maltose	Lactose	Salicin	Arabinose	Xylose	Fructose	Galactose	Rhamnose	Sorbitol	Inulin	Dextrin	Inositol	Raffinose	Dulcitol	Trehalose	Glycogen	Penicillin	
<i>Bacteroides fragilis</i> <sup>a,h</sup> <i>B. pneumoniae</i> <sup>a,h</sup> <i>B. putidus</i> <sup>e,f</sup> <i>B. funduliformis</i> <sup>b,d-j</sup> <i>B. serpens</i> <sup>e,k,l</sup> <i>B. nigrescens</i> <sup>g,h,m,n</sup>	0	0	V	V	V	V	V	0	+	0 <sub>v</sub>	4.6 - 5.4	AC <sub>v</sub>	±	A	A	V	0 <sub>v</sub>	A	A <sub>v</sub>	A <sub>v</sub>	0 <sub>v</sub>	0 <sub>v</sub>	A <sub>v</sub>	A	A	0 <sub>v</sub>	V	A	0	A	0	0 <sub>v</sub>	A <sub>v</sub>	R	
	0	0			0	0	±	0		5.5	5.5		0	A	0	0	0	A	A	A	0	A	A	A	0	0	A	0	0	0	0	0	0		
	0		0	f	V	+	+		+			P <sub>v</sub>	0	0	0	0	0	0							0 <sub>v</sub>	0	0	0	V	0	V	0	0 <sub>v</sub>	0 <sub>v</sub>	V
	0	+	0 <sub>v</sub>	xf	0	+	+	+	0 <sub>v</sub>	+	+	5.6 - 6.5	A <sub>v</sub>	+	A	A <sub>v</sub>	0 <sub>v</sub>	0 <sub>v</sub>	A	A <sub>v</sub>	V	0 <sub>v</sub>	V	A	A <sub>v</sub>	0 <sub>v</sub>	0	0	V	0	V	0	0 <sub>v</sub>	0 <sub>v</sub>	V
	+	0	0	0	f <sub>v</sub>	+	+	+		+	+		AC	+	A	0	0	0	A	A	0	0	A	A	A		0	0	A	0	A	0	0	S	
<i>Fusobacterium fusiforme</i> <sup>f-h,p-r</sup> <i>F. girans</i> <sup>f,k,t</sup>	0	0 <sub>v</sub>	0	f	0	+	+	0 <sub>v</sub>	+	0	6.0 - 6.9	0	V	A	0 <sub>v</sub>	0	0	0 <sub>v</sub>	0	0	0	A*	A <sub>v</sub>	0 <sub>v</sub>	0	0	0	0	0	0	0	0	0	0	S
	+		x	x	0	0	0 <sub>v</sub>	0 <sub>v</sub>	+	0	6.2 - 6.9	AC	+	A	A	V	V	A	A	A	A	A	A	A											
<i>Vibrio sputorum</i> <sup>r,t,u</sup> <i>Spirillum putigenum</i> <sup>r,t,v</sup>	+	a		0	0	0	f <sub>v</sub>	V	+			0	0	0	0	0				0	0										0			S	
	+			0	0	0	0	+			5.1 - 5.4	AC	0	A	A	A <sub>v</sub>			A																

V, variable; A, acid; C, clot; P, peptonized; f, foul; x, acid; a, green; R, resistant; S, sensitive; 0, negative.

\* Based on 1 strain, reference h

a Eggerth and Gagnon (1933)

b Henthorne et al. (1936)

c Weiss and Reiger (1937)

d Smith and Ropes (1947)

e Prevot (1957)

f Beerens (1953-54)

g Garrod (1955)

h Sonnenwirth (1960)

i Lohelle (1947)

j Dack et al. (1937, 1938)

k Prevot (1938)

l Steen and Thodea (1950)

m Oliver and Wherry (1921)

n Schwabacher et al. (1947)

o Burdon (1932)

p Poe (1941)

q Boyer (1956)

r Rosebury et al. (1950)

s Prevot (1940)

t Macdonald (1953)

u Moore (1954)

v Macdonald et al. (1959)

\* Rosebury, Theodor: Microorganisms Indigenous To Man. The Blackiston Division, McGraw-Hill Book Company, Inc., New York, N.Y., pp. 150-151, 1962.

TABLE 35. CLASSIFICATION OF FA AND GD TYPES

CATENABACTERIUM  
FA-1 (*C. cateniforme*)  
GD-5

RAMIBACTERIUM  
FA-9 (*R. pleuriticum*)  
FA-17 (*R. ramosum*)

FUSOBACTERIUM  
FA-3  
FA-18  
GD-1  
GD-2  
GD-7

SPHAEROPHORUS  
FA-2  
FA-10  
FA-16  
GD-4 (*S. necrophorus*)

EUBACTERIUM  
FA-4  
FA-6  
FA-11  
FA-12

BACTEROIDES  
FA-7  
FA-15  
GD-3 (*B. putidus*)  
GD-6 (*B. funduliformis*)

VEILLONELLA  
FA-13

LACTOBACILLUS  
FA-5 (*L. bifidus*)

BUTYRIBACTERIUM  
FA-14 (*B. rettgeri*)

DIALISTER  
FA-8

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14. KEY WORDS	LINK A		LINK B		LINK C	
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